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STUDIES ON MONOGENETIC TREMATODES

III. REDESCRIPTIONS AND VARIATIONS IN KNOWN SPECIES[1]

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The primary purpose of this paper is (1) to present more complete descriptions of some known TETRAONCHINAE, (2) to record structural and size variations that occur under different environmental conditions, and (3) to record new host and/or geographical records for previously described species. Accomplishment of these objectives has been made possible by means of material collected from Tennessee waters by Dr. R. V. Bangham of Wooster College, Wooster, Ohio.

The present writer, in 1936, was impressed by the fact that specimens of *Urocleidus mucronatus* (Mizelle, 1936) from the bluegill sunfish (*Lepomis macrochirus*) from Lake Senachwine, Henry, Illinois, were nearly twice the size of those taken from the same host at Lake Decatur, Decatur, Illinois. Mueller (1937) described *U. contortus* as being similar to *U. principalis* (Mizelle, 1936) except for the fact that the distal prongs (two) of the accessory piece were not confluent as described for the latter species. Both conditions with reference to this structure, were reported (Mizelle, 1938) for *U. principalis,* and *U. contortus* was reduced to synonymy. Seamster (1938) found such wide variations in the anchors of *Cleidodiscus pricei* Mueller, 1936, that they were offered as partial reason for reduction of the genus *Haplocleidus* Mueller, 1937, to a synonym (in part) of *Urocleidus* Mueller, 1934, (Mizelle & Hughes, 1938).

Size variations in *U. mucronatus* from the two Illinois localities mentioned above, are considered valid since they concern adult forms taken from the same host species, during the same season, and treated similarly with reference to methods of technique. In the present investigation, variations reported for chitinous parts as anchors, bars, hooks, cirri, and accessory pieces are also interpreted as valid since these structures undergo no apparent shrinkage during fixation. Variations in soft parts, however, may not be exactly as reported because of (1) a difference in

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killing and fixation methods used by different men and (2) a difference in the number of specimens used as bases for measurements. Species described by the present author (1936, 1938) were relaxed before fixation and measuring, while the present material was not relaxed but was field-fixed with formalin. Mizelle (1936, 1938) used twenty specimens as a basis for description of each species. Present material permits use of thirteen or fewer specimens for each species. Since there is no statement to the contrary it is assumed that Mueller (1934, 1936, 1937) used fixed material (unrelaxed). This writer did not indicate how many specimens were used in the determination of his measurements.

It is thought by the present writer that a better picture of a species is presented if several specimens serve as a basis for description. To augment this would entail use of several specimens from different hosts.

Quotations found in the redescriptions are those available for comparison from Mueller's papers (1934, 1936, 1937). See Mizelle and Seamster (1939) for methods employed in this work.

The author wishes to thank Dr. Bangham again for the opportunity to examine this material.

Urocleidus principalis (Mizelle, 1936) Mizelle & Hughes, 1938
Figs. 17–29

Hosts and Localities: Kentucky or Spotted Bass, *Micropterus punctulatus* (Rafinesque), Cove Creek, Caryville, Tenn.; Norris Lake, Norris, Tenn.; and Cold Creek, below Norris Dam (Tenn.). Small-Mouth Bass, *Micropterus dolomieu* Lacépède, Cove Creek, Caryville, Tenn. Large-Mouth Bass, *Huro salmoides* (Lacépède), Hatchery, Norris, Tenn.

Previously Reported Hosts and Localities: Kentucky or Spotted Bass, Salt Fork of the Big Vermilion River, Homer, Ill. (Mizelle, 1936). Small-Mouth Bass, Boomer Creek, Stillwater, Okla. (Seamster, 1938) and Salt Fork of the Big Vermilion River, Homer, Ill. (Mizelle, 1938). Large-Mouth Bass, Fla. (Mueller, 1937); Baton Rouge, La. (Summers & Bennett, 1938); and Lake Senachwine, Henry, Ill. (Mizelle, 1938).

Synonyms: Onchocleidus principalis Mizelle, 1936, and *Onchocleidus contortus* Mueller, 1937.

Site of Infestation: Gills.

Number of Specimens used in this Investigation: Ten.

The specimens used in this investigation were five from the Kentucky bass, four from the large-mouth bass, and one (the only one available) from the small-mouth bass. For all measurements (Table 1) *U. principalis* from the Kentucky bass possessed minima and maxima for cirrus length and dorsal anchor width. Maxima for these structures were shared by one parasite each from the small-mouth (cirrus) and large-mouth bass (dorsal anchor width). In addition forms from the Kentucky bass possessed four minimum measurements (dorsal bar length, ventral anchor width, dorsal anchor length, and hook length) and five maximum measurements (body length, pharynx diameter, haptoral length and width, and accessory piece length). The ventral anchor

minimum width was shared by a parasite from the large-mouth bass. Specimens from the large-mouth bass possessed minimal and maximal measurements for body width, ventral bar length, and ventral anchor width. The minimum for the last-named structure was shared by a parasite from the Kentucky bass. Forms from the large-mouth bass furnished also, two minimum measurements (haptoral length, and accessory piece length) and five maxima (dorsal bar length, ventral and dorsal anchor length, dorsal anchor width, and hook length). The one available parasite from the small-mouth bass possessed four minimal measurements (body length, pharynx diameter, haptoral width, and ventral anchor length) and one maximum measurement (cirrus length) which was shared by a parasite from the Kentucky bass. From the foregoing it is clear that no one of these three bass hosts from this region permit maximum growth of all parasite structures considered in this work. Instead, the minima and maxima are rather well distributed among the parasites from all three hosts (Table 1).

With reference to variation in specimens from different parts of the country it was found that northern forms were larger than those from Tennessee for all average measurements (composite) except body width (equal), ventral anchor length (longer in southern forms), width and length of dorsal anchors (longer in Tenn. forms, widths equal), and cirrus length (longer in Tenn. forms) (Table 2). The pharynx in Tennessee specimens was either longer than broad or vice versa. The bars of Illinois forms are comparatively regular in shape (Mizelle, 1938, Figs. 83, 84) while those in Tennessee specimens exhibit striking variations (Figs. 19, 20, 22–25, 27, 29). An apron-like projection on the ventral bar as described by Mueller (1937) has not been observed by the present author. Specimens of this parasite from the large-mouth bass nearly always have the anchor shafts (especially those of the ventrals) bent in the direction of the anchor points (Figs. 26, 28) whereas those from the other two bass hosts possess much straighter anchor shafts (Mizelle, 1938, Figs. 76, 77). One specimen from the large-mouth bass presented ventral anchors which were dissimilar in the shape of the bases (Figs. 18, 21). Haptoral hooks of southern (Tenn.) forms are slightly shorter than those of northern specimens but the shapes are the same. Mueller (1937) reported hooks in Florida specimens to be equal in length (0.02 mm) with pair number five (seven of Mueller) "slightly smaller than others."[2] The present writer has invariably found differences in length of the various haptoral hooks (Mizelle, 1938 and Table 1 and 2). Members of hook pairs numbers one and five are shorter than the rest of the hooks (Mizelle, 1938). The cirrus length in Florida forms was reported by Mueller to be 0.030 mm which is shorter than the minimal length found by the present author for Illinois and Tennessee

[2] Quotations in the redescriptions are from Mueller's papers (1934, 1936, 1937).

specimens. As previously reported the accessory piece may exist with a distal fork or with the prongs confluent to form a ring through which the cirrus passes (Mueller, 1937, Fig. 15; Mizelle, 1938, Figs. 81, 82). For the sake of comparison, it is unfortunate that Mueller (1937) gave only five measurements in his description. He did not indicate how many specimens furnished a basis for each of these. Comparisons of measurements for forms from the Kentucky bass from Illinois and Tennessee (Tables 1 and 2) present a picture similar to that for the Kentucky bass from Illinois and for the three previously named basses (composite measurements) from Tennessee, except that forms from the latter state possess a shorter body width and a cirrus length equal to that of the northern forms.

Urocleidus furcatus (Mueller, 1937) Mizelle & Hughes, 1938

Hosts and Localities: Kentucky or Spotted Bass, *Micropterus punctulatus* (Rafinesque), Norris Lake, Norris, Tenn. and Cold Creek, below Norris Dam (Tenn.). Large-Mouth Bass, *Huro salmoides* (Lacépède), Hatchery, Norris, Tenn.

Previously Reported Host and Localities: Large-Mouth Bass, Fla. (various localities) (Mueller, 1937) and Baton Rouge, La. (Summers & Bennett, 1938).

Synonym: Haplocleidus furcatus Mueller, 1937.

Site of Infestation: Gills.

Number of Specimens used in this Investigation: Ten.

Redescription: Small dactylogyrids usually with a smooth cuticula. Length 0.352 mm (0.238–0.495 mm), greatest body width 0.107 mm (0.081–0.135 mm). Eye spots of posterior pair larger and invariably closer together than those of the anterior pair. Cephalic lobes well defined. Pharynx ovate in outline, usually longer than wide but may be wider than long, and often rotated from the normal alignment; transverse diameter 0.025 mm (0.022–0.030 mm). Haptor well set off from the body and roughly pentagonal in outline, length 0.067 mm (0.058–0.082 mm), width 0.082 mm (0.068–0.091 mm). Bars dissimilar in shape; dorsal with knobbed ends, ventral with a median posterior projection. Length of ventral bar 0.028 mm (0.025–0.032 mm), length of dorsal bar 0.030 mm (0.020–0.037 mm). Anchors dissimilar in shape and size (dorsals larger). Wings on ventral anchors well defined, but low and inconspicuous on dorsals. Solid anchor shafts and points junction to form an internal angle. Superficial root of each dorsal anchor base markedly elongate and curved in direction of anchor point, deep root degenerate; dorsal anchor length 0.074 mm (0.043–0.081 mm), "0.069 mm.," width 0.015 mm (0.014–0.016 mm). Ventral anchors with slightly bifurcate or nonbifurcate bases, length 0.035 mm (0.025–0.038 mm), "0.035 mm.," width 0.015 mm (0.011–0.018 mm). Hooks normal with reference to number and arrangement. Each hook with an ovate to narrowly elliptical base, a slender solid shaft, a sickle-shaped termination, and an opposable piece; lengths 0.011–0.026 mm. Hooks of pairs numbers one and five somewhat shorter than the others which are of about the same length. Testis ovate and lies posterior to the ovary which is ovate to circular in outline. Vagina dextral. Copulatory complex consists of a cirrus, cirral thread, and accessory piece. Cirrus a straight or undulate, chitinous tapered tube with a comparatively small base and pointed distal extremity; length 0.064 mm (0.062–0.066 mm), "0.049 mm." Cirral thread coiled about the cirrus. Accessory piece composed of a rod with a forked termination, the prongs of which may be confluent to form a ring through which the cirrus passes; accessory piece length 0.032 mm (0.020–0.038 mm), "0.023 mm." Seminal vesicle large, elliptical in outline and often containing spermatozoa. Vitellaria moderately developed.

Available specimens (1) from Kentucky bass and measurements (4) from Florida specimens (Mueller, 1937) are insufficient in number to

permit valid comparisons of specimens from different hosts and localities (Table 3).

Actinocleidus fusiformis (Mueller, 1934) Mueller, 1937

Figs. 1–6

Hosts and Locality: Kentucky or Spotted Bass, *Micropterus punctulatus* (Rafinesque), Norris Lake, Norris, Tenn.; Cold Creek, below Norris Dam (Tenn.); and Cove Creek, Caryville, Tenn. Small-Mouth Bass, *Micropterus dolomieu* Lacépède, Cove Creek, Caryville, Tenn. Large-Mouth Bass, *Huro salmoides* (Lacépède), Hatchery, Norris, Tenn.

Previously Reported Hosts and Localities: Small-Mouth Bass, State Hatchery Reservoir, Constantia, N. Y. (Mueller, 1934) and Ohio Hatcheries, London, Ohio (Mueller, 1936). Large-Mouth Bass, Peace River, Fla. (Mueller, 1937); Boomer Creek, Stillwater, Okla. (Seamster, 1938); and New Roads, La. (Summers & Bennett, 1938).

Synonyms: Ancyrocephalus cruciatus (Wedl.). See Cooper (1915). *Cleidodiscus fusiformis* Mueller, 1934.

Site of Infestation: Gills.

Number of Specimens used in this Investigation: Thirteen.

Redescription: The earliest record for this parasite in North America is indicated by a figure for *Ancyrocephalus cruciatus* (Wedl., 1857) by Cooper (1915). Shape, exclusive of haptor, narrowly elliptical in dorsal view. Length 0.440 mm (0.270–0.910 mm), "0.550 mm. to 0.850 mm.," greatest body width 0.149 mm (0.068–0.190 mm), "0.170 mm." Pharynx may be either longer than wide or *vice versa*, transverse diameter 0.042 mm (0.029–0.059 mm), "0.065 mm." Anterior eye spots smaller and closer together than members of the posterior pair. Cephalic lobes poorly developed or wanting. Haptor disc-like, transverse diameter 0.099 mm (0.082–0.116 mm), "about 0.096 mm.," and set off from the body by a comparatively narrow peduncle. Anchor shafts (anterior generally solid, posterior hollow) and points regularly recurved, bases deeply bifurcate, wings well defined. Length of anterior anchors 0.041 mm (0.039–0.046 mm), width 0.018 mm (0.014–0.026 mm). Length of posterior anchors 0.047 mm (0.043–0.053 mm), width 0.018 mm (0.012–0.025 mm). "The four main hooks are of approximately the same size and shape . . . greatest dimension of a hook . . . is 0.044 mm. . . . width of shaft at the broadest point is 0.0136 mm." Fourteen haptoral hooks (arrangement as shown by Mueller, 1936, Fig. 8), each consisting of a small oval base, a slender shaft, a sickle-shaped termination, and an opposable piece; lengths 0.011–0.019 mm. Anterior bar V-shaped or may exist with the arms of the V depressed, length 0.058 mm (0.049–0.068 mm); posterior bar modified to form a plate to which the anterior bar articulates, length (in reality the width of the plate at its widest part) 0.042 mm (0.035–0.047 mm). As shown in Fig. 4, the central portion of the plate may be absent. Vagina located in anterior body half on left margin; seminal receptacle prominent. Ovary ovoid or reniform, about the same size as, or slightly larger than, testis and situated with the long axis at right angles, or obliquely to, the body axis. Testis ovoid, located posterior, posterodorsal, or dorsal, to the ovary and with greatest diameter parallel or transverse to the body axis. Seminal vesicle and prostate prominent; cirrus as described by Mueller (1936), length 0.051 mm (0.038–0.057 mm), "0.04 mm." Accessory piece about same length as, and generally articulated to, the cirrus. Balsam and glycerine-jelly mounts often show the accessory piece disarticulated from the cirrus (Figs. 5, 6). Mueller's figures (1936, Figs. 15, 16) show this structure approximately one-half the length of the cirrus. Vitellaria moderately developed and extend into the tapered portion of the posterior body half.

Comparative measurements for this parasite from the three bass hosts (Tenn.) present much the same picture as that for *Urocleidus principalis* in that the minima and maxima are rather well distributed among forms

from all three hosts. Size differences in northern and southern forms are shown in Table 4.

New host records for this species are interesting since Mueller (1936) recovered it often enough from the small-mouth bass (only) to believe it apparently specific for this host.

Actinocleidus bursatus (Mueller, 1936) Mueller, 1937

Hosts and Locality: Kentucky or Spotted Bass, *Micropterus punctulatus* (Rafinesque), and Small-Mouth Bass, *Micropterus dolomieu* Lacépède, Cove Creek, Caryville, Tenn.

Previously Reported Host and Localities: Large-Mouth Bass, *Huro salmoides* (Lacépède), London, Ohio (Mueller, 1936) and Peace River, Fla. (Mueller, 1937).

Synonym: Ancyrocephalus bursatus Mueller, 1936.

Site of Infestation: Gills.

Number of Specimens used in this Investigation: Ten.

Redescription: Comparatively large robust parasites with thick transversely wrinkled cuticula. Body length 0.820 mm (0.646–1.006 mm), width 0.328 mm (0.172–0.405 mm). Cephalic lobes poorly developed or wanting. Eye spots of anterior pair smaller and may be closer together or farther apart than those of the posterior pair. Pharynx broader than long, transverse diameter 0.111 mm (0.061–0.130 mm); "about 0.08 mm. in diameter." Haptor consisting of (1) a central pedicellate portion bearing anchors, bars, and hooks numbers one and five and (2) a surrounding thin subcircular scalloped-margined portion. Each marginal indentation of the haptor is near a hook, except the median anterior and posterior clefts which are very marked and cut the haptor almost to the pedicellate portion. Haptor broader than long, transverse diameter 0.242 mm (0.134–0.294 mm), "about 0.2 mm. in diameter," length 0.216 mm (0.144–0.270 mm). Anterior bar bent posteriorly in midportion, length 0.031 mm (0.026–0.036 mm), "about 0.04 mm. long." Posterior bar articulate to the posterior margin of the anterior bar and modified to form a plate-like structure with an anterior median notch and two posterior arms. Posterior bar length 0.019 mm (0.015–0.022 mm), "about 0.025 mm. × 0.01 mm." Anchors similar in shape and size, bases bifurcate, superficial roots longer than deep roots. Hollow anchor shafts and solid points unite with a suggestion of an internal angle. Length of anterior anchors 0.027 mm (0.025–0.028 mm), width 0.011 mm (0.008–0.013 mm). Length of posterior anchors 0.027 mm (0.022–0.030 mm), width 0.011 mm (0.009–0.013 mm). "Anchors rather similar in size and shape, about 0.03 mm. long, not following the curve." Anchor wings poorly developed and apparently wanting in many specimens. Hooks (14) similar in shape and size; each consisting of a subspherical base, a comparatively long slender shaft, a sickle-shaped termination with a posterior projecting spike, and an opposable piece; lengths 0.014–0.021 mm. Mueller (1936, Figs. 6, 7) represents these structures as elongate rods with slightly curved distal extremities. Hook arrangement departs from that of most Tetraonchinae. Hook pair number one lies immediately anterior to the anterior anchors, number five is situated between the two pairs of anchors, each of the remaining hooks located on the haptoral margin near an indentation. Gonads comparatively small, spherical to ovate in shape. When ovate they may lie with the long axis parallel, transverse, or diagonal to, the median line. Testis smaller than, and situated posteriorly or dorsoposteriorly to, and often with the anterior border overlapping the posterior margin of, the ovary. Seminal vesicle large, copulatory complex well developed. Cirrus as drawn by Mueller (1936, Fig. 20), length 0.041 mm (0.039–0.046 mm). Accessory piece length 0.027 mm (0.025–0.028 mm), "Accessory piece present, about half the length of the cirrus." Vagina located on left body margin near the mid-length; seminal receptacle not observed. Vitellaria composed of heavy numerous follicles.

Only six measurements (matching) are available from Mueller's description so that very slight comparison can be made for north and

south forms. Of these measurements, three are less and three are greater than those (averages) for southern specimens (Table 5). Minimum and maximum measurements in this work are shared by specimens from the two bass hosts.

Actinocleidus articularis (Mizelle, 1936) Mueller, 1937

Host and Locality: Long-Eared Sunfish, *Lepomis megalotis* (Rafinesque), Cove Creek, Caryville, Tenn.

Previously Reported Host and Locality: Long-Eared Sunfish, Embarrass River, Urbana, Ill. (Mizelle, 1936).

Synonym: Cleidodiscus articularis Mizelle, 1936.

Site of Infestation: Gills.

Number of Specimens used in this Investigation: Three.

Tennessee specimens of this species conform to the general morphology given in the original description. All average measurements, except body length, pharynx diameter, and haptoral length, are as great or greater than, those for Illinois specimens (Table 6). The author regrets that more specimens of this parasite were not available from Tennessee.

Cleidodiscus banghami (Mueller, 1936) Mizelle, 1940

Figs. 7–16

Hosts and Locality: Kentucky or Spotted Bass, *Micropterus punctulatus* (Rafinesque), and Small-Mouth Bass, *Micropterus dolomieu* Lacépède, Cove Creek, Caryville, Tenn.

Previously Reported Host and Locality: Small-Mouth Bass, London, Ohio (Mueller, 1936).

Synonyms: Tetracleidus banghami Mueller, 1936, and *Urocleidus banghami* (Mueller, 1936) Mizelle & Hughes, 1938.

Site of Infestation: Gills.

Number of Specimens used in this Investigation: Ten.

Redescription: Body robust or at times with posterior portion drawn out to form an elongate peduncle to which the haptor is attached. Cephalic lobes wanting, cuticula thick and transversely wrinkled. Body length 0.566 mm (0.405–0.691 mm), "about 0.4–0.5 mm. long," greatest body width 0.273 mm (0.198–0.328 mm), "0.17 mm. in diameter." Anterior cephalic region usually with a median cleft. Eye spots of anterior pair smaller and invariably farther apart than those of the posterior pair. Pharynx usually longer than wide but may be wider than long, transverse diameter 0.057 mm (0.040–0.069 mm), "about 0.07 mm." Haptor knoblike, relatively small, and much shorter than wide; width 0.101 mm (0.089–0.115 mm). "Haptor small, about 0.06 mm. long, by 0.13 mm. wide." Bars slightly bent in midportions and approximately equal in length. Ventral bar usually with two short projections on the posterior border (Figs. 14, 15), length 0.036 mm (0.032–0.045 mm). Dorsal bar more massive and usually with three blunt projections on the posterior margin but may exist as a comparatively smooth structure (Figs. 11–13), length 0.036 mm (0.030–0.046 mm). "Supporting bars of about the same size and shape, 0.04 mm. in length." Anchors similar in shape, wings scarcely evident on most specimens. Ventral anchor bases slightly bifurcate or nonbifurcate, dorsal anchor bases slightly bifurcate. Anchor shafts and points hollow and regularly recurved. Length of ventral anchors 0.027 mm (0.025–0.029 mm), width 0.021 mm (0.018–0.024 mm). Length of dorsal anchors 0.025 mm (0.022–0.026 mm), width 0.017 mm (0.015–0.019 mm). "Largest dimension of anchor, not following the curve, 0.034 mm." Hooks normal in number and distribution, subequal in size. Each hook consists of a very small base (scarcely evident in many cases), a slender shaft, a sickle-shaped termination, and an opposable piece; lengths 0.012–0.015 mm. Gonads near midportion of

body. Ovary transversely elongate, ovate to elliptical in outline; much smaller than, and with posterior margin covered by, the testis. Testis transversely or longitudinally elongate, roughly ovate in outline, and from two to several times as large as the ovary. Vagina dextral; seminal receptacle large. Copulatory complex consisting of a cirrus and an accessory piece which are basally articulate with each other. Cirrus a large thin chitinous tube often with a solid basal posterior-projecting structure (Fig. 8), cirrus length 0.068 mm (0.058–0.079 mm), "about 0.07 mm. long." Accessory piece an elongate chitinous rod with a retrorse tip, length 0.065 mm (0.053–0.071 mm), "about 0.06 mm. long."

This species was originally placed in the now obsolete genus *Tetracleidus* by Mueller (1936). Mizelle and Hughes (1938) in a review of the subfamily TETRAONCHINAE removed it to *Urocleidus* Mueller, 1936, on the basis of a dextral vagina and nonarticulate cirrus and accessory piece. Mueller's specimen (used for comparison) and his figure (1936, Fig. 33) show these structures to be nonarticulate. This was probably due to pressure applied at the time of preparation of the material used. Some of the writer's specimens present this nonarticulate condition but in such forms a well-defined articulating surface is present on each structure (Figs. 7, 8). Other specimens show these structures basally articulate (Figs. 9, 10) and on this basis this species is transferred to the genus *Cleidodiscus* Mueller, 1934. *C. banghami* is the only species in this genus to possess a dextral vagina. This condition is considered insufficient to reinstate the genus *Tetracleidus* since species of *Urocleidus* may or may not possess a vagina. Comparisons of specimens from Ohio and Tennessee are interesting since southern forms are larger (average) than northern specimens. The converse of this is true for *U. principalis, Actinocleidus fusiformis,* and *A. articularis.* See Tables 2, 4, and 7 for these, and Table 7 for comparisons of structures of *C. banghami.*

Cleidodiscus bedardi Mizelle, 1936

Host and Locality: Long-Eared Sunfish, *Lepomis megalotis* (Rafinesque), Cove Creek, Caryville, Tenn.

Previously Reported Host and Locality: Long-Eared Sunfish, Embarrass River, Urbana, Ill. (Mizelle, 1936).

Site of Infestation: Gills.

Number of Specimens used in this Investigation: One.

All measurements, except body width (0.051 mm), for this single specimen were between the minima and maxima given in the original description (Mizelle, 1936). Nine available specimens from the type locality yielded a cirrus measurement of 0.030 mm (0.024–0.033 mm) for length and an accessory piece length of 0.029 mm (0.022–0.032 mm).

SUMMARY

Material collected from Tennessee by Dr. R. V. Bangham of Wooster College, Wooster, Ohio, has rendered possible the redescriptions of *Actinocleidus fusiformis, A. bursatus, Cleidodiscus banghami,* and *Uro-*

cleidus furcatus. These have been made for each species on the basis of ten or thirteen specimens taken from two or more hosts. New geographical and host records, or new geographical records only, have been made for the foregoing and other species, namely, *Cleidodiscus bedardi, Actinocleidus articularis,* and *Urocleidus principalis.* Comparisons of body dimensions and critical internal structures have been made for each species with reference to hosts and localities. These are presented by descriptions and/or tables of measurements. In most cases, parasites from Illinois localities possess greater body size but do not always have larger internal structures than specimens from Tennessee. Minima and maxima for parasites of a given species (from Tenn.) are rather uniformly distributed among specimens from the available hosts.

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Explanation of Tables

AP Accessory piece
A Anterior
AL Anterior bar length
C Cirrus
D Dorsal
DL Dorsal bar length
HD Haptoral diameter (transverse)
H Hooks
L Length
Ph Pharynx (transverse diameter)
P Posterior
PL Posterior bar length
V Ventral
VL Ventral bar length
W Width

Measurements are given in microns. Bold face figures indicate minima and maxima for parasites from Tennessee hosts. Italicized figures indicate exceptions listed in the text.

TABLE 1.—*Measurements of* U. principalis *from Three Bass Hosts (Tenn.)*

	L	W	Ph	Haptor L	Haptor W	Bars VL	Bars DL	Anchors V L	Anchors V W	Anchors D L	Anchors D W	H	C	AP	
Min.	199	86	19	43	71	28	**31**	35	**14**	**25**	**12**	**11**	**32**	20	Kentucky Bass Host, 5 Specimens
Max.	**329**	111	**24**	**62**	**110**	36	35	44	19	37	**17**	22	**49**	**28**	
Ave.	252	95	23	50	81	32	32	38	16	32	15		40	23	
Min.	207	**83**	19	**37**	67	**27**	32	38	**14**	32	14	12	37	**19**	Large-Mouth Bass Host, 4 Specimens
Max.	289	**117**	22	58	73	**40**	**38**	**46**	**23**	**41**	**17**	**24**	45	26	
Ave.	237	97	20	47	71	33	36	42	19	38	15		41	23	
——	**189**	104	**18**	38	**54**	32	33	**33**	15	26	13	13–22	**49**	27	Small-Mouth Bass Host, 1 Specimen

TABLE 2.—*Measurements of* U. principalis *from Different Hosts and Localities*

	L	W	Ph	Haptor L	Haptor W	Bars VL	Bars DL	Anchors V L	Anchors V W	Anchors D L	Anchors D W	H	C	AP	
Min.	369	70	25	51	78	32	30	28	15	27	13	13	36	22	Kentucky Bass Host, Illinois Locality, 20 Specimens* (Mizelle, 1938)
Max.	631	127	34	82	125	42	44	38	21	34	17	25	47	27	
Ave.	467	*96*	29	66	98	36	35	*33*	18	*31*	*15*		*40*	25	
Min.	189	83	18	37	54	27	31	33	14	25	12	11	32	19	Kentucky, Large-Mouth, and Small-Mouth Bass Hosts, Tennessee Locality, 10 Specimens
Max.	329	117	24	62	110	40	38	46	23	41	17	24	49	28	
Ave.	240	*96*	21	48	75	33	34	*39*	17	*34*	*15*		*41*	24	
——								44**		39**		20	30	23	Small-Mouth Bass Host, Florida Locality, ? Specimens (Mueller, 1937)

* The cirrus and accessory piece measurements have been supplemented through use of ten and six specimens (respectively) from the type locality.

** Mueller (1937) did not state which of these measurements was for dorsal or ventral anchors.

TABLE 3.—*Measurements of* U. furcatus *from Different Hosts and Localities*

	L	W	Ph	Haptor L	Haptor W	Bars VL	Bars DL	Anchors V L	Anchors V W	Anchors D L	Anchors D W	H	C	AP	
Min.	**238**	**81**	**22**	**58**	**68**	**25**	**20**	**25**	**11**	**43**	**14**	**11**	**62**	**20**	Large-Mouth Bass Host, Tennessee Locality, 9 Specimens
Max.	**495**	**135**	**30**	**82**	86	**32**	**37**	**38**	**18**	**81**	**16**	**26**	**66**	**38**	
Ave.	350	112	26	67	79	28	32	36	16	75	15		64	33	
——	360	**81**	23	61	**91**	**32**	**20**	**25**	**11**	**43**	**16**	13–22	64	22	Kentucky Bass Host, Tennessee Locality, 1 Specimen
Ave.	352	107	25	67	82	28	30	35	15	74	15		64	32	Composite Average for Two Above-Named Hosts
——								35		69			49	23	Large-Mouth Bass Host, Florida Locality, ? Number of Specimens (Mueller, 1937)

TABLE 4.—*Measurements of* A. fusiformis *from Different Hosts and Localities*

	L	W	Ph	HD	Bars AL	Bars PL	Anchors A L	Anchors A W	Anchors P L	Anchors P W	H	C	
Min.	380	123	42	**82**	51	**35**	**39**	**14**	**43**	**12**	14	48	Small-Mouth Bass Host, Tennessee Locality, 4 Specimens
Max.	477	161	**59**	115	54	40	42	15	49	18	**19**	55	
Ave.	430	149	47	102	53	38	40		46	14		51	
Min.	**270**	**68**	**29**	85	59	45	40	19	44	18	**11**	**38**	Large-Mouth Bass Host, Tennessee Locality, 4 Specimens
Max.	**910**	180	43	97	**68**	**47**	43	**26**	**53**	**25**	18	55	
Ave.	483	132	35	89	64	46	42	22	51	21		49	
Min.	360	108	43	95	**49**	39	40	**14**	**43**	18	15	49	Kentucky Bass Host, Tennessee Locality, 5 Specimens
Max.	495	**190**	46	**116**	67	46	**46**	21	47	19	18	**57**	
Ave.	412	163	45	107	58	42	42	17	45			54	
Ave.	440	149	42	99	58	42	41	18	47	18		51	Composite Average for the Three Above Hosts
——	550–850	170	65	96			44	13.6*				40	Small-Mouth Bass Host, New York & Ohio Localities,** ? No. of Specimens (Mueller, 1934 & 1936)

* Anchors were reported by Mueller (1934) to be of approximately equal size.
** Cirrus length is the only measurement given for New York forms.

TABLE 5.—*Measurements of* A. bursatus *from Different Hosts and Localities*

	L	W	Ph	Haptor L	Haptor W	Bars AL	Bars PL	Anchors A L	Anchors A W	Anchors P L	Anchors P W	H	C	AP	
Min.	675	**172**	**61**	178	**134**	28	16	26	9	23	**9**	**14**	**39**	26	Spotted Bass Host, Tennessee Locality, 3 Specimens
Max.	920	**405**	120	200	225	34	21	**28**	**13**	27	**13**	19	**46**	**28**	
Ave.	802	320	91	195	169	30	19	27	11	27	11		43	27	
Min.	**646**	276	97	**144**	259	**26**	**15**	**25**	**8**	**22**	**9**	**14**	**39**	**25**	Small-Mouth Bass Host, Tennessee Locality, 7 Specimens
Max.	**1006**	383	**130**	**270**	**294**	**36**	**22**	**28**	13	**30**	13	**21**	41	**28**	
Ave.	828	331	115	220	279	32	19	26	11	27	11		40	27	
Ave.	820	328	111	216	242	31	19	27	11	27	11		41	27	Composite Average for the Two Above Hosts
——			80		200	40	25	30*					30		Large-Mouth Bass Host, Ohio Locality, ? No. of Specimens (Mueller, 1936)

* The two pairs of anchors were reported by Mueller (1936) to be of similar size.

TABLE 6.—*Measurements of* A. articularis *from Different Localities*

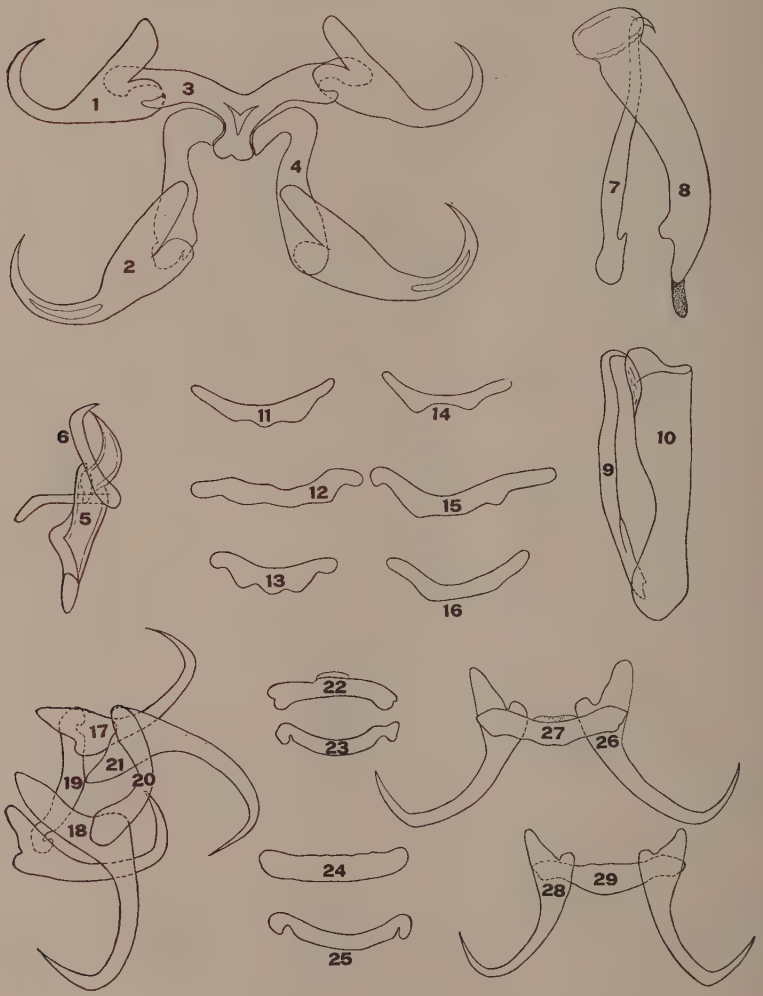
	L	W	Ph	Haptor L	Haptor W	Bars AL	Bars PL	Anchors A L	Anchors A W	Anchors P L	Anchors P W	H	C	AP	
Min.	360	49	21	47	61	32	19	32	9	28	8	9	29	15	Long-Eared Sunfish Host, Illinois Locality, 20 Specimens* (Mizelle, 1938)
Max.	615	79	28	57	86	42	25	38	13	38	11	19	31	17	
Ave.	*470*	68	*25*	*55*	72	37	22	33	10	32	10		30	15	
Min.	256	80	19	49	71	41	26	35	9	34	9	9	29	14	Long-Eared Sunfish Host, Tennessee Locality, 3 Specimens
Max.	270	86	24	57	75	44	28	37	11	36	11	19	35	17	
Ave.	*261*	84	*21*	*52*	73	42	27	36	10	35	10		32	15	

* Measurements for cirrus and accessory piece have been supplemented through the use of five specimens from the type locality. The accessory piece length was made from the base to the end of the pointed distal extremity.

TABLE 7.—*Measurements of* C. banghami *from Different Hosts and Localities*

	L	W	Ph	HD	Bars VL	Bars DL	Anchors V L	Anchors V W	Anchors D L	Anchors D W	H	C	AP	
Min.	**405**	**198**	**40**	90	34	**30**	27	**18**	23	16	**12**	**58**	**53**	Small-Mouth Bass Host, Tennessee Locality, 5 Specimens
Max.	599	**328**	**69**	**115**	**45**	**46**	**29**	22	**26**	18	15	71	**71**	
Ave.	502	260	56	103	37	37	28	20	25	17		65	62	
Min.	545	265	48	**89**	**32**	33	**25**	21	**22**	**15**	**12**	62	64	Kentucky Bass Host, Tennessee Locality, 5 Specimens
Max.	**691**	315	64	102	38	39	28	**24**	**26**	**19**	15	**79**	69	
Ave.	630	287	59	98	35	36	27	23	24	17		70	67	
Ave.	566	273	57	101	36	36	27	21	25	17		68	65	Composite Average for the Two Above Hosts
	400–500	170	70	130	40	40	34*					70	60	Small-Mouth Bass Host, Ohio Locality, ? No. of Specimens (Mueller, 1936)

* This measurement is given by Mueller (1936) as the greatest dimension of an anchor. No mention is made as to the one for which it is intended.



EXPLANATION OF PLATE

All figures were made at the same magnification with the aid of a camera lucida.

Actinocleidus fusiformis (Mueller, 1934) Mueller, 1937

FIG. 1. Anterior anchor
FIG. 2. Posterior anchor
FIG. 3. Anterior bar
FIG. 4. Posterior bar
FIG. 5. Cirrus
FIG. 6. Accessory piece

Cleidodiscus banghami (Mueller, 1936) Mizelle, 1940

FIGS. 7 & 9. Accessory pieces
FIGS. 8 & 10. Cirri
FIGS. 11–13. Dorsal bars
FIGS. 14–16. Ventral bars

Urocleidus principalis (Mizelle, 1936) Mizelle & Hughes, 1938

FIGS. 17, 28. Dorsal anchors
FIGS. 18, 21, 26. Ventral anchors
FIGS. 19, 23, 25, 29. Dorsal bars
FIGS. 20, 22, 24, 27. Ventral bars

A NEW HUMAN HELMINTHIC INFECTION IN CUBA

Pedro Kourí, M.D.
with the collaboration of
Irving Rappaport, M.S.[1]

Since 1935 we have found in eleven human cases a species of Cyclophyllidea not previously reported in Cuba. After studying the first few specimens, we thought that we had found a new species of *Raillietina*.[2] Later, with the kind cooperation of Professor J. G. Baer, study of new material containing scoleces caused us to rectify our first impression. According to Professor Baer, this species, possibly new, must be placed in the Genus *Inermicapsifer* Janicki, 1910; Subfamily Linstowinae Fuhrmann, 1907; Family Anoplocephalidae, Fuhrmann 1907.

The characteristics of this cestode are as follows:

It is 27–42 cm long, possessing 310–368 proglottids. The scolex measures about 630 micra long by 610 micra broad. The suckers are about 185 micra in the broadest diameter and 150 micra deep. The neck is roughly 3 mm long. The proglottids in the midportion of the strobila are broader than long, approximately 2.3 by 1.5 mm. Gravid proglottids expelled in the feces measure from 3 to 3.75 mm in length, by 1 to 2 mm in breadth. In the living motile condition, one of the poles of the freed proglottid is often broader, thus giving the segment a club-shaped appearance.

Genital pores are unilateral and in the center of the margin of each mature proglottid. The cirrus pouch measures about 150 micra long by 75 micra broad. The pouch is small and runs centrally and somewhat anteriorly. The vas deferens is long and coiled both without and within the cirrus pouch. The cirrus is often seen projecting from the genital pore. Testes number from 33 to 49 per segment, the antiporal side possessing, in general, about 5 more testes than the poral side, due to the presence of the vagina and vas deferens in the latter side. The testes average 65 by 72 micra. Gravid segments contain between 48 and 175 egg capsules. These capsules are very variable in size and shape. Each contains from 6 to 11 ova. These eggs are 55 by 49 micra and contain a small hexacanth embryo, 22 by 19 micra. Hooklets of the embryo are about 6 micra long.

Within the past 6 months we have been led to believe that the incidence of this species in Cuba is much greater than was heretofore suspected. The first case was brought to our attention in 1935 and two more in 1937. A report of this parasite was published and widely distributed among Cuban physicians in June, 1938. From January, 1939, until October, we have had no less than 8 cases brought to our attention, thus bringing the total up to 11.[3]

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[1] From the Department of Tropical Medicine and Parasitology, University of Havana, and Department of Public Health and Preventive Medicine, Cornell University Medical College, New York City. Read before Section V of the 3rd International Congress for Microbiology, New York City, September 2–9, 1939.

[2] Abstracts of Communications, 3rd Internat. Cong. Microbiol., p. 176.

[3] The number of cases reported up to now (March, 1940) totals 18.

In distribution, one case is from the province of Pinar del Rio, and one from the province of Matanzas. The other nine cases are all from Havana Province. Three of the nine cases occurred within a distance of one kilometer of one another. In fact, two of the children parasitized lived next door to each other.

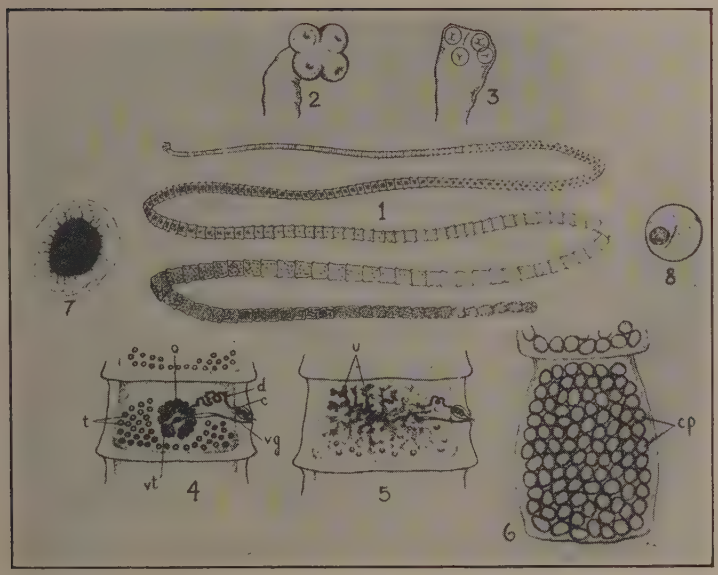
Nine of the cases probably possessed one parasite each, the others 2, 2 and 3, respectively. According to the mother of one of the parasitized children, gravid proglottids had been expelled continuously for 6 years, except for one interruption lasting 4 months. The age of the human host thus far has been found to vary from 13 months to three and one-half years, with the exception of the nine-year-old boy who apparently has been expelling segments for 6 years.

Parasitism is usually discovered by the child's parents, who notice small whitish bodies resembling rice grains in the stools. Occasionally fragments of strobilae are expelled. Expulsion of mature proglottids in stools is regular, about 3 to 6 daily for each parasite harbored. Microscopic examination of feces is negative for eggs, but macroscopic examination and screening reveal mature proglottids. In fresh feces, the gravid proglottids are found to migrate to the surface.

The parasite induces slight or no symptoms. Expulsion of the complete worm follows treatment with ether extract of male fern and carbon tetrachloride. Even coconut oil may be effective in expulsion of fragments or of an entire worm. This suggests that the parasite is not a very resistant one in the human intestine though the case reported in this paper which was apparently parasitized for six years, if true, would lead one to believe otherwise.

It is yet unknown how infection occurs, whether the parasite has any habitual definitive host other than man,[4] or whether development is direct or indirect.

[4] Both Dr. Baer and Dr. Stunkard suggested that this parasite may not be a true human parasite and believe that the rat may be the definitive host. We hold the same opinion but about 100 rats examined were all found negative for this worm.



Inermicapsifer sp. from Cuba.

1. Entire specimen.
2. Retracted scolex of the same specimen.
3. Not retracted scolex of another specimen.
4. Mature segment, with (t) testes; (d) vas deferens; (c) cirrus pouch; (o) ovary; (vt) vitellaria; (vg) vagina.
5. Proglottid in a more advanced stage of development with ramified uterus (u).
6. Gravid proglottid full of egg capsules (cp).
7. Egg capsule.
8. Egg.

ABSENCE OF VITELLINE MEMBRANES ON DEVELOPING EGGS IN PARASITIC FEMALES OF *STRONGYLOIDES RATTI*[1]

B. G. Chitwood[2] and G. L. Graham

The alternative modes of larval development which occur in *Strongyloides ratti* Sandground, 1925, and apparently in all other species of the genus *Strongyloides* as well, have stimulated considerable investigation concerning the biological factors involved in producing this rather unusual phenomenon. In spite of the definitive points which have been established, resolution of the problem has been impeded, and clarification, even for *S. ratti,* is only at the penultimate stage.

Let us review the life cycle of *S. ratti* briefly. The parasite is normally found embedded in the mucosa of the small intestine in rats. Notably, only females have ever been observed. The eggs shed by the parasite complete their development in a fecal medium outside the body of the host. Some of the young larvae metamorphose directly into infective filariform larvae; others develop into morphologically distinct, sexually differentiated, free-living males and females which then produce a crop of infective filariform larvae morphologically indistinguishable from those that develop directly from the eggs of the parasite. These two modes of larval development are known as homogonic, or direct; and heterogonic, or indirect, respectively.

In its fundamental aspects the problem is essentially simple: How can one account for the production of two types of progeny by a *single* gynomorphic parasite, in one type of which sexual differentiation must also be accounted for? Confronted thus with the problem of determining whether parasitic *S. ratti* females reproduced parthenogenetically or syngonically, the writers, in addition to searching for hermaphroditically produced spermatozoa in the genital tubes of the parasite, have sought for evidence of fertilization in the eggs of this parasite.

MATERIALS AND METHODS

Strongyloides ratti parasites from two pure lines with distinctive biological potentialities were used. In one instance the parasites were from stock infections which had been built up from a single, heterogonically derived parasite through the course of several generations by mass infections. The pure indirect line of single *S. ratti* from which these mass

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infections were derived was shown by Graham (1939) to produce predominantly progeny of heterogonic development, approximately 90 per cent of the total progeny being free-living males and females. On the other hand, the other line of *S. ratti* used in this study had been built up to a mass level from a pure direct line of singly established *S. ratti.* In this line (direct line of Graham, 1939, and direct line I of Graham, 1940a) only about 15 per cent of the total progeny were free-living males and females. Established at mass levels, these lines of *S. ratti* continued to show their respective characteristics; one yielded predominantly homogonic larvae, the other predominantly heterogonic adults.

Free-living females from the bisexual generation in the heterogonic line were also studied. These were used after they had developed in fecal cultures from 30 to 36 hours and 48 hours. Both fertilized and unfertilized eggs from free-living *S. ratti* females were also utilized in testing the reaction of reagents on shells and vitelline membranes.

A number of parasitic females of *Strongyloides* sp. from the common gray squirrel, *Sciurus carolinensis,* were also studied. Eggs from these parasites were likewise tested with reagents.

As comparative check material in the study of the developing eggs of *S. ratti,* eggs of the root knot nematode, *Heterodera marioni,* were employed. *H. marioni* eggs possess a readily observed vitelline membrane. Their reaction to reagents was reported by Chitwood (1938).

RESULTS

Observations were made on sealed saline mounts of parasitic *Strongyloides ratti* females with the specific objective of demonstrating spermatozoa in this reputedly syngonic form. In all instances parasites from 5 to 10 days old were studied. Being young, they were favorable material. Examinations were made immediately following their recovery from the intestine of the rat host. All were producing eggs in abundance and these were developing in an apparently normal manner. Using optical equipment which revealed details of structure with great clarity, prolonged search of the genital tubes of both intact worms and worms from which the internal organs had been extruded by cutting off the tail failed to reveal sperm cells.

Collateral observations made the absence of spermatozoa in these parasites assume a significant aspect. Polar bodies were observed in a number of eggs, but the majority of eggs examined did not show them. When they were present, usually only one was seen in any egg. In several instances the polar body was at the distal end of the egg, but they were as frequently found at the proximal end of the egg or even in a lateral position. The usual pattern in nematode eggs which are fertilized is for more than one polar body to be thrown out, the first representing the meiotic division, being opposite the point of sperm entry,

i.e., at the distal end of the egg. In the few instances in which two polar bodies were seen in an egg, they were always lying side by side, thus creating the impression that they had been formed by a division rather than representing the extrusion of two polar bodies from the egg. Walton (1918) has indicated that in *Toxocara canis* eggs rotation of the cytoplasm within the fertilization membrane may make it appear that the second polar body is given off at another position on the surface of the egg. While these observations in themselves are questionable evidence of non-fertilization, the random positions in which singly occurring polar bodies were seen support such a conception.

In one case the polar body was clearly resting against the egg shell. Ordinarily in fertilized nematode eggs, a fertilization or vitelline membrane is present and polar bodies are within, not outside, this membrane. Hence the observation of a polar body resting against the egg shell suggested the probable absence of such a membrane, unless it were closely adherent to the shell. Tests with various reagents failed to reveal a vitelline membrane. Eggs were rapidly stained with dilute iodine and with dilute gentian violet. In the presence of a vitelline membrane, penetration and staining of the egg mass would have been markedly delayed. Following treatment of the eggs with absolute alcohol (which is a solvent for the lipoidal vitelline membrane of nematode eggs), staining with iodine and gentian violet was not hastened. Heating also had no effect. This treatment melts and thus destroys the protective lipoidal vitelline membrane, permitting the entry of dyes. When "Clorox" (a 5 per cent solution of sodium hypochlorite which is a solvent for protein and chitin) was applied, the entire egg, including the shell, disappeared without leaving a trace of a vitelline membrane, which would have been undamaged by this reagent.

On the other hand, eggs of *Heterodera marioni* with a clearly visible vitelline membrane did not show staining of the embryo when treated with gentian violet. When treated with "Clorox" the shell was rapidly dissolved, but the embryo was protected by the vitelline membrane; however, when the "Clorox" was washed out with water and the shell-free eggs were treated with absolute alcohol, the vitelline membrane was quickly dissolved and the embryo freed. When eggs of *H. marioni* with shells were treated with absolute alcohol, then washed with water, and treated with dilute gentian violet, the embryo was quickly stained by the dye, indicating that the membrane had been removed by the alcohol.

Although the presence of spermatozoa had been anticipated at the outset of these observations on parasitic *S. ratti* females (in part because of the rarity of parthenogenesis in nematodes and also because Sandground, 1926, had reported the presence of spermatozoa in parasitic *S. ratti* females), a thorough search failed to reveal them in this favor-

able material. The additional evidence that spermatozoa had not been present—namely, the observation of developing eggs in which a vitelline membrane could not be seen directly and no evidence of this membrane's presence found even when reagents were employed—indicated that these eggs were developing parthenogenetically. It is important to note that no distinction could be discerned either in parasites or in eggs from them irrespective of whether they were from a line producing a high percentage of free-living adults among their progeny or whether they were from the line producing predominantly direct development larvae.

On the other hand, examination of females from the free-living bisexual generation of *S. ratti* presented an entirely different picture. Feces were collected from rats harboring massive infections of *S. ratti* from a line producing predominantly adults of the heterogonic generation. Females isolated from fecal cultures incubated for 30 to 36 hours were studied. In many of them, numerous sperm cells were readily observed in the genital tubes at the junction of the uterus proper and the ovary. Polar bodies were at the distal end of developing eggs, not at the proximal end, i.e., the originally attached end, which is the characteristic, though not invariable, point of sperm entrance in bisexually reproducing nematodes. The vitelline membrane was easily seen even without the use of reagents. The membrane was destroyed by heating and treatment with absolute alcohol; eggs thus treated were rapidly stained by iodine and gentian violet, whereas the membrane in untreated eggs inhibited the entrance of these dyes. Treatment with "Clorox" dissolved the egg shell rapidly but left the vitelline membrane intact. This disappeared when it was heated or treated with absolute alcohol. Thus the tests with developing eggs from *S. ratti* females of the free-living bisexual generation were comparable to those with eggs of the root knot nematode, *H. marioni,* which also possess a vitelline membrane.

In those free-living females where sperm cells were not demonstrable (virgin or senescent individuals) no development of the eggs was seen and neither polar bodies nor vitelline membrane could be demonstrated by direct observation. Even when reagents were used for the latter the results were negative. Thus the contrast between the condition in parasitic *S. ratti* females and *S. ratti* females from the free-living bisexual generation was remarkably distinct. Morphologically the detail of the genital tubes in these two types of *S. ratti* females is very similar. Reproductively they seem to pursue distinctly different lines. The free-living females follow the familiar nematode pattern, whereas the parasitic females, insofar as these observations can determine, are unusual in that their eggs possess no vitelline membrane and appear to develop parthenogenetically.

Studies of parasitic females of *Strongyloides* sp. from the gray squirrel, *Sciurus carolinensis,* were comparable in every respect to those

made on *S. ratti.* No spermatozoa were seen in the genital tubes, although granules and cell clusters which might be mistaken for sperm cells were observed. Developing eggs did not possess a directly observable vitelline membrane nor could such membrane be demonstrated with the use of reagents. Polar bodies when present were usually single and were in random positions. In the few instances when two were observed in any egg they were always adjacent to each other. Apparently, parthenogenesis also occurs in the eggs of parasitic females of this form. Free-living females from the bisexual generation were not studied.

Unpublished observations by one of us (B. G. C.) indicate that the free-living nema, *Rhabditis filiformis,* for which only one sex is known, also reproduces by parthenogenesis. The developing eggs of this worm possess no vitelline membrane. To the writers' knowledge, these are the only known cases where evidence has been offered that parthenogenesis occurs in nematodes in the absence of spermatozoa, although it seems predictable that similar instances can be multiplied. The nearest approach to these cases is afforded by the free-living nematode, *Rhabditis aberrans,* in which parthenogenesis occurred even though sperm cells were present. Krüger (1913) observed insemination of ova in this protandrous hermaphrodite, but the male pronucleus did not fuse with the female pronucleus; instead it degenerated in the egg plasm without taking part in the formation of the embryonic nucleus. Under such conditions, only one polar body was produced. Apparently the only function of the spermatozoon—essential though it was for the development of the egg—was the initiation of cleavage. It should be noted that Krüger also observed *R. aberrans* to produce a very small percentage of male worms in cultures. Admitting the purity of the specific strain of the nematode with which she worked, the presence of a few males and the observation of a single egg in which two polar bodies and fusion of the male and female pronuclei were noted, suggest that parthenogenesis is not an exclusive feature in the reproduction of *R. aberrans.*

DISCUSSION

Certain historical aspects of the controversy concerning the reproductive status of parasitic females of various species of *Strongyloides* have been admirably presented and discussed by Sandground (1926). As he pointed out, Rovelli (1888) appears to have been the first investigator to examine parasitic *Strongyloides* females from the rat and subsequently report that they reproduced parthenogenetically. Unfortunately Rovelli's paper was inaccessible to Sandground and he depended on the accuracy and completeness of a review of it by Braun (1888). Granting this premise, Sandground's disposal of the reported evidence in favor of parthenogenesis was entirely justifiable. On the

other hand, examination of the article by Rovelli leads to an impression different from that gained from Braun's review alone and forces one to keep in mind the possibility that parthenogenesis may occur in *Strongyloides* even if Rovelli's observations are not full proof thereof.

Rovelli (1888) stated that he made observations "sullo *Strongyloides* del *Mus decumanus* e su quello della Pecora"; not on "die Arten aus der Wanderratte und dem Schweine" as reported by Braun (1888). He noted further that there "resulterà chiaro dall'esame dell'apparato genitale (dalla sua origine fino alla estremità terminale, dalla iniziale formazione delle ova fino al loro completo sviluppo), non havvi giammai neppure un lontano sospetto della presenza degli elementi fecondatori maschili, e perciò mi credo autorizzato ad affermare (come incliniva ad ammettere il GRASSI e contrariamente alle asserzioni del LEUCKART) che *gli Strongyloides sono indubbiamente femmine partenogenetiche allo stadio di parassiti, mancando affatto il ricettacolo del seme e qualsiasi traccia di spermatozoi.*" (Italicized in the original.) Whatever deficiencies there may be in Rovelli's interpretation of his further observations, the statement above is clear and definite. While "this evidence is all of a negative character" as Sandground (1926) stated, it does not necessarily follow that "its reliability is not substantial." The writers believe that the absence of spermatozoa in parasitic *S. ratti* females studied by them is a "substantial" factual confirmation of Rovelli's observation, further fortified by the absence of vitelline membranes on the developing eggs.

It was in a definite attempt to account for the alternative modes of larval development on a "germinal chromosome basis" that Sandground (1926) "decided to re-investigate the sexual condition of the parasitic generation of Strongyloides." As he observed (p. 375), "In Strongyloides, the well established belief that the parasitic generation is represented by females whose eggs develop parthenogenetically, has in the past automatically precluded the possibility of explaining the mechanism that determines the dual mode of development that this organism displays on the basis of an heterozygosis." Conceding the objectivity of his study in the course of which he examined "living specimens of *S. ratti*" and found spermatozoa "in appreciable numbers" at the point where the egg passes from the oviduct into the uterus, his observations stand in contrast to those in the present case in which no spermatozoa were encountered in any of the many worms examined. It is, of course, conceivable that he may have been dealing with a syngonic strain of the parasite differing from the parthenogenetic strains used in the present study. On the other hand, Sandground illustrated his report with photomicrographs (p. 348) of stained longitudinal sections of parasitic *S. ratti*. The caption of one of these states, "Shows oocytes and sperm

cells; the tissue on the right of the section is packed with sperm cells, but at this focus they are only seen on the periphery." The section appears to have been cut somewhat obliquely and the cells at the periphery bear a striking resemblance to those of the uterine epithelium. Under such circumstances, it seems possible that the portion of the section which was "packed with sperm cells" may represent a frontal section of the cells of the uterine wall and not spermatozoa as he supposed.

Although we do not deny that Sandground (1926) may have seen spermatozoa in the living material of *S. ratti* which he examined, the present study of living *S. ratti* parasites failed to confirm the point. To the extent that his reproduced photomicrographs are fair objective evidence of what he found, his general conclusions in the matter must be considered with caution.

While the writers agree with Sandground (1926) that easy acceptance of the idea of parthenogenesis on the part of many previous authors was unfortunate and unjustified on a factual basis, it is equally unjustifiable to assume that it cannot occur. Thus Leuckart (1879) when confronted with "unverkennbare Samenkörperchen" in the uterus of *Ascaris nigrovenosa* [= *Rhabdias bufonis*], a monoecious parasite in the lungs of frogs, observed, "Ich muss übrigens hinzufügen, dass ich in manchen Fällen—und ebenso ist es auch andern Helminthologen ergangen (nach brieflicher Mittheilung z. B. v. Siebold)—vergebens nach Samenkörperchen gesucht habe, die Möglichkeit einer parthenogenetischen Entwicklung also noch keineswegs vollständig ausschliessen möchte."

The direct morphological evidence of parthenogenesis in parasitic *S. ratti* females herein reported is compatible with the observations made by one of us (G. L. G.) on pure lines of singly established *S. ratti*. Constitutionally distinct lines of this parasite have been demonstrated (Graham, 1939; 1940a; 1940b). In those lines which were maintained by serial, homogonic passage (single larva infections), no evidence of constitutional deviation has ever been observed. On the other hand, serial passage in a heterogonically maintained line of singly established *S. ratti* has been shown to produce parasites on several occasions with biological and morphological characteristics which are constitutionally dissimilar from those of the parent from which they were derived. When these new lines with distinctive characteristics were maintained by serial homogonic passage, no discernible change was ever observed in succeeding generations of the parasite. These are facts which bear directly on the problem of determining the type of egg development in *S. ratti* of the parasitic generation. They provide no evidence that the parasite possesses a mechanism for inheritance of characteristics comparable to that observed in the free-living bisexual generation. Thus they obviously

indicate the absence of males, at least functional males, in the parasitic generation and also deny the occurrence of hermaphroditic heterozygosis.

SUMMARY

Parasitic *Strongyloides ratti* females from a pure line producing predominantly progeny of homogonic development as well as from another pure line producing predominantly progeny of heterogonic development have been studied. In both cases, spermatozoa were not demonstrable in the genital tubes of these gynomorphic parasites nor could the presence of a vitelline membrane (evidence of fertilization) be shown in developing eggs of these parasites either by direct observation or the use of reagents.

On the other hand, sperm cells could be seen in free-living *S. ratti* females of the heterogonic generation and vitelline membranes seen in developing eggs as well as demonstrated by tests with reagents.

These results indicate that female *S. ratti* in the parasitic generation reproduce parthenogenetically whereas normal bisexual reproduction occurs in the free-living generation. Examination of parasitic females of *Strongyloides* sp. from the gray squirrel, *Sciurus carolinensis*, indicated that these parasites are reproductively comparable to parasitic *S. ratti* females, i.e., parthenogenetic.

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NOTES ON JUVENILE ACANTHOCEPHALA

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Most of the juvenile Acanthocephala that have been described are marine or exotic species. The present paper adds two species to the list of those known to occur in North American fresh-water hosts.

Pomphorhynchus bulbocolli Linkins, 1919

While examining pike from the Great Lakes a number of small fish which could not be identified were found in the pike stomachs. These small fish were then examined, and an amphipod containing a juvenile specimen of *Pomphorhynchus bulbocolli* was recovered from one of them. This specimen (Fig. 1) is a female and its measurements are as follows: Body 4.9 mm long by 0.7 mm in maximum diameter. Proboscis 0.4 by 0.2 mm. Neck 1.3 by 0.23 mm. Proboscis hooks in 12 longitudinal rows with 14 hooks each. Anterior hooks 0.04 mm long and posterior hooks 0.02 mm long. The ovary has dissociated to form four masses about 0.07 by 0.05 mm located in the ligament.

The mesenteries of these smaller fish contained numerous specimens of encysted acanthocephalans which could be identified as *P. bulbocolli* by the number and arrangement of proboscis hooks. The size of the cysts (Fig. 2) varied from 0.7 by 0.3 mm to 2 by 0.4 mm. The worms seemed to have degenerated and very little of their internal structure could be distinguished. Meyer (1933) suggests that acanthocephalan cysts in the second or third intermediate host undergo degeneration after reaching a certain age, or degenerate as a result of defense mechanisms of the host and improper nourishment.

Adults of *P. bulbocolli* have been reported from a number of host species including *Catostomus commersonii, Cyprinus carpio, Erimyzon sucetta oblongus* and *Notemigonus crysoleucas.* Van Cleave (1934) states that the suckers and sucker-like fishes are the most frequent hosts of this species. Cysts of *P. bulbocolli* have been reported from *Percina caprodes,* although adult specimens were found in the intestine of this host at the same time (Van Cleave, 1919). No record of the occurrence of this species in an arthropod has been found in the literature.

Leuckart (1862) found juvenile stages of *Echinorhynchus proteus* (*sic*) in the body cavity of *Gammarus pulex.* He was probably dealing with *Pomphorhynchus laevis* or *Polymorphus minutus,* as suggested by Meyer (1933). Leuckart also conducted an experiment in which he infected *Gammarus* with the eggs of this species and obtained juvenile

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forms of the parasite. Von Linstow (1892) found *Pomphorhynchus laevis* encysted in small fish, and thought that they served as the first intermediate host. He observed the parasites in *Gammarus,* but believed that this was an abnormal occurrence. Riquier (1909) fed *Tinca vulgaris* containing encysted juvenile forms of *P. laevis* to pike, *Esox lucius,* in which the parasites developed to maturity.

Centrorhynchus sp.

Eleven encysted juvenile acanthocephalans have been recovered from the intestinal wall of a water snake, *Natrix sipedon,* obtained from Kentucky by Dr. R. M. Cable. These specimens belong to the genus *Centrorhynchus* Lühe, 1911, adults of which occur in birds. The male specimens (Fig. 4) from the water snake vary in length from 3.4 to 4.2 mm and the average maximum diameter of the body is 0.58 mm. The females are slightly larger. The proboscis varies in length from 0.94 to 1.1 mm and in diameter from 0.28 to 0.3 mm. It is armed with 28 to 33 longitudinal rows of hooks (Fig. 3), each row containing 18 to 22 hooks. The first four hooks measure 0.05 to 0.06 mm in length and possess roots having an average length of 0.07 mm. The six hooks posterior to these have an average length of 0.04 mm and bear small roots. The remaining spines have an average length of 0.03 mm and do not bear roots. The proboscis sheath has an average length of 0.86 mm. The ribbon-like lemnisci usually measure 0.7 mm in length. The reproductive organs are not well developed. In the males the widely separated testes are ovoid with an average size of 0.07 by 0.06 mm; the tubular cement glands usually measure 0.36 by 0.07 mm. In the females the ligament extends from the proboscis sheath to the posterior end of the body; the gonads could not be observed.

Although the diagnostic features of the specimens of *Centrorhynchus* sp. from the water snake do not correspond to those of any species described in the literature, it does not seem desirable to establish a new species at this time since these specimens are immature and the adult stage is unknown. Two species of *Centrorhynchus* have been described from the United States. Van Cleave (1924) reported adults of *C. spinosus* from a bird, *Herodias egretta,* and Millzner (1924) reported juvenile forms of *C. californicus* from *Hyla regilla.* The writer's specimens have a distinctly larger proboscis than that of *C. spinosus,* and the proboscis hooks are much more numerous in *C. californicus* than in the specimens reported here. Of the 30 species which have been reported from other localities many of them are incompletely described; hence, it is difficult to compare the writer's specimens with these species.

The final host of the specimens from the water snake is probably a predacious bird. The osprey (*Pandion haliaetus*) has been seen at the

locality from which the water snake was taken, but the writer has had no opportunity to examine these birds for parasites. Since the first intermediate host of ACANTHOCEPHALA is almost always an arthropod, the role of the water snake is probably that of a second intermediate host.

A number of European and South American species of *Centrorhynchus* have been reported from amphibians and reptiles. Mingazzini (1896) obtained cysts of *C. aluconis* and *C. buteonis* from *Zamenis gemonensis* and fed them to nestlings of *Falco tinnunculus*. Some of the parasites became established in the intestine, others escaped with the feces of the host, while still others became encysted in the body cavity. In a study of ACANTHOCEPHALA from Japan, Van Cleave (1925) found immature individuals of *Centrorhynchus* sp. encysted in the intestinal wall of *Rana rugosa* and the body wall of *R. japonica*.

SUMMARY

Juvenile *Pomphorhynchus bulbocolli* have been found encysted in the mesenteries of small fish, and a specimen was recovered from an amphipod in the stomach of one fish.

A juvenile acanthocephalan belonging to the genus *Centrorhynchus* is reported from the intestinal wall of the water snake, *Natrix sipedon*.

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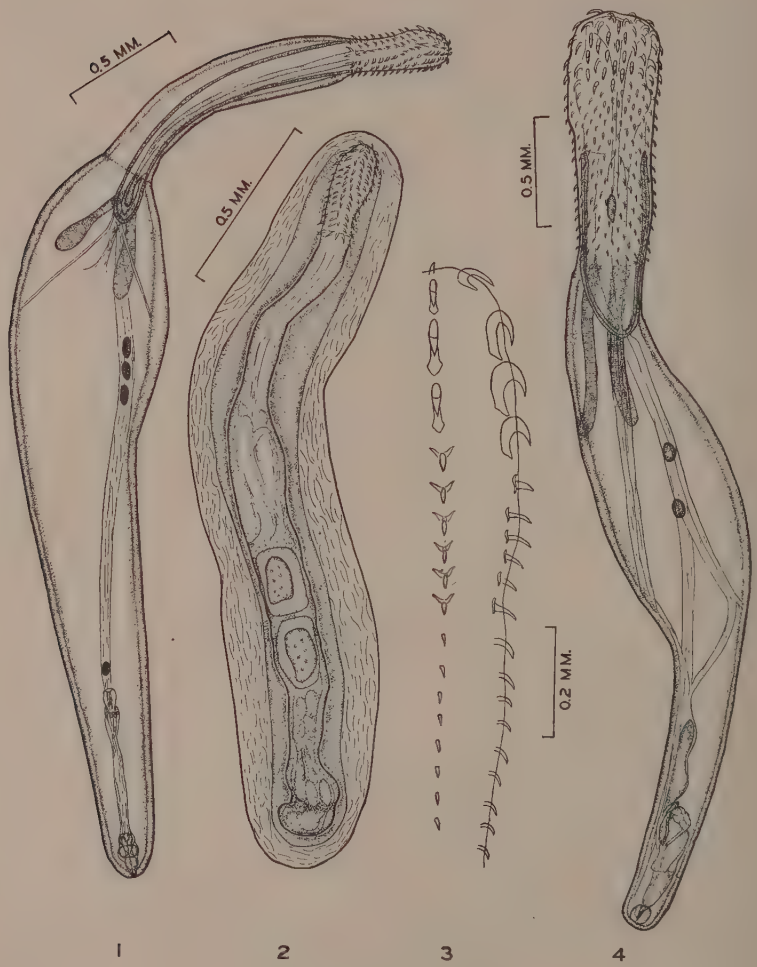

EXPLANATION OF PLATE

All drawings were made with the aid of a camera lucida.

FIG. 1. *Pomphorhynchus bulbocolli,* juvenile female from amphipod.

FIG. 2. Encysted *P. bulbocolli,* juvenile male from mesentery of fish.

FIG. 3. Surface and profile views of two rows of hooks enlarged from the specimen shown in Fig. 4.

FIG. 4. *Centrorhynchus* sp., juvenile male from cyst in intestinal wall of water snake.

TWO NEW SPECIES OF SCHISTOSOME CERCARIAE FROM WISCONSIN[1]

STERLING BRACKETT

In a program of investigation of schistosome dermatitis in Wisconsin a large number of snails have been collected from lakes and streams throughout the state and examined for trematode infections. Many of these collections were made specifically to determine the species of snails which harbor the schistosome cercariae with which we are already acquainted and to attempt to discover new species of these cercariae. Only two previously undescribed schistosome cercariae have been found thus far. These both occur in the snail species *Gyraulus parvus* (Say) and have been found only in a single locality, a small vegetation filled lake (to be referred to as the Sun Prairie pond) fifteen miles north of Madison, Wisconsin. For these two cercariae, which are here described as new species, the names *Cercaria elongata* n. sp. and *C. gyrauli* n. sp. are proposed. It is very easy to distinguish these two species from one another both by their activity and by their general appearance under the dissecting microscope. This is fortunate in view of the fact that both occur in the same host and could easily become mixed during the studies.

The methods employed were essentially those outlined by Talbot (1936) in his studies of some dermatitis-producing schistosome cercariae. Certain minor modifications were necessary because of the small size of the snail host and the fact that relatively few cercariae were shed into the water each day by infected snails.

Cercaria elongata n. sp.

(Fig. 2)

Specific diagnosis: Schistosome cercaria. Entire body, tail stem and furcae uniformly spined. Body length[2] 198–270 μ, mean 228 ± 3.1 μ; distance from posterior end of body to middle of ventral sucker 81–117 μ, mean 101 ± 1.4 μ; body width 40–56 μ, mean 46 ± 1.2 μ; tail stem length 396–522 μ, mean 459 ± 6.3 μ; tail stem width 30–40 μ, mean 34 ± 0.6 μ; furcal length 252–306 μ, mean 279 ± 2.6 μ, oral sucker length 60–92 μ, mean 72 ± 1.7 μ; oral sucker width 28–44 μ, mean 36 ± 0.7 μ; ventral sucker width 16–36 μ, mean 21 ± 0.6 μ. Ventral sucker only slightly protruded. Dorso-ventral furcal fin fold present. Five pairs of penetration glands in emerged cercariae, three large and two small ducts leading forward and opening through spines. Digestive system a long tubular esophagus bifurcating just in

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[1] A contribution from the Department of Zoology of the University of Wisconsin. Appreciation is expressed for the encouragement and help received from Dr. C. A. Herrick. This work was aided by a grant from the Wisconsin Alumni Research Foundation.

[2] From measurements of 28 specimens killed in three groups (10, 10 and 8) according to the Talbot method (Talbot, 1936). The extremes and the probable error of the mean are used to indicate the character of the variation.

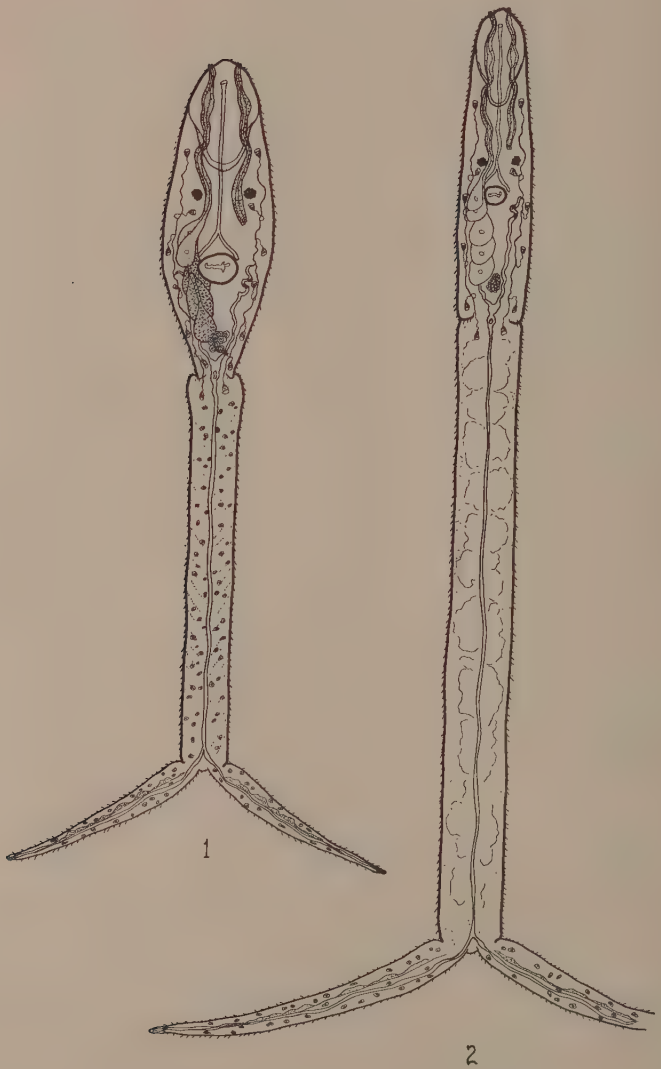

FIG. 1. Semidiagrammatic drawing of *C. gyrauli* showing structural details, ventral view.

FIG. 2. Semidiagrammatic drawing of *C. elongata* showing structural details, ventral view.

Both figures drawn approximately to the same scale.

front of ventral sucker into two short ceca. Excretory system six pairs of flame cells, 2[(2) + (3+1)], five in body, two anterior and three posterior, and one in anterior part of tail stem. Anterior and posterior collecting tubules join bladder arms on a level with middle of ventral sucker. Bladder arms with several loose loops close to junction with collecting tubules and each with two ciliated patches in the looped section.

Host: Gyraulus parvus (Say).

Locality: Sun Prairie, Dane County, Wisconsin, U. S. A.

The presence of only five flame cells on each side of the body of *C. elongata* clearly separates it from *C. elvae* Miller, 1923, *C. stagnicolae* Talbot, 1936, *C. physellae* Talbot, 1936, *C. ocellata* LaVal., 1855, the cercaria of *Bilharziella polonica* (*cf.* Szidat, 1929) and the cercaria of *B. yokogawai* Oiso, 1927, all of which have six flame cells on each side of the body except the last which has seven. In addition it has a shorter body and longer tail stem than any of the above species. The cercaria of *Schistosomatium douthitti* and *C. tuckerensis* Miller, 1927, possess the same number of flame cells in the body as *C. elongata* but while they have three flame cells on the anterior and two on the posterior collecting tube *C. elongata* has the condition reversed with only two on the anterior and three on the posterior collecting tube. In addition, *C. elongata* differs distinctly from these two in size and proportions. Even under the lower powers of magnification of the dissecting microscope *C. elongata* can be quite easily distinguished because of its extremely long and slender proportions and by the fact that the tail stem is over twice as long as the body.

In its swimming activities *C. elongata* might be confused only with *C. stagnicolae* since it tends to hang motionless in the water on the side of the container towards the light in a manner similar to *C. stagnicolae* which was described by Cort and Talbot (1936). With experience, the size difference is noticeable and since the snail hosts are so different no confusion should arise.

The ventral sucker is small and inconspicuous and not protruded as it usually is in *C. elvae* and its relatives. The ventral sucker is likewise situated proportionately farther forward than it is in the "elvae" group. The eye spots are situated in a position very close to the anterior margin of the ventral sucker. Vital stains used by Talbot in his studies on some of the dermatitis-producing cercariae were not taken up readily by the glands of *C. elongata*. The ducts leading forward were more easily studied and the exact number readily determined. There are three larger and two smaller ducts in a group on each side. Only freely escaped cercariae were studied so escape glands were not seen although from inference they undoubtedly occur. A suggestion of the presence of weakly outlined caudal bodies in the tail stem similar to those described for many strigeid cercariae was noted. The sporocysts were not studied in detail

since all of the material available was used in attempts to determine the complete life cycle of this species.

Cercaria gyrauli n. sp.
(Fig. 1)

Specific diagnosis: Schistosome cercaria. Entire body, tail stem and furcae uniformly spined. Body length[3] 198–270 μ, mean 215 ± 3.3 μ; body width 56–100 μ, mean 73 ± 1.9 μ; distance from middle of ventral sucker to posterior end of body 54–90 μ, mean 73 ± 1.8 μ; tail stem length 270–342 μ, mean 298 ± 2.8 μ; tail stem width 24–36 μ, mean 29 ± 0.3 μ; furcal length 126–162 μ, mean 135 ± 1.5 μ; length of oral sucker 60–92 μ, mean 77 ± 1.6 μ; width of oral sucker (12 specimens) 36–60 μ, mean 46 ± 1.9 μ; diameter of ventral sucker 24–32 μ, mean 28 ± 0.5 μ. Ventral sucker only slightly protruded. Dorso-ventral furcal fin fold present. Five pairs of penetration glands in emerged cercariae, three large and two small ducts leading forward and opening through spines. Genital primordium a group of 4–8 cells with large nuclei close to posterior end of body. Digestive system a long tubular esophagus bifurcating in front of ventral sucker into two short ceca. Excretory system seven pairs of flame cells, 2[(3) + (3+1)], six in body and one in anterior portion of tail stem. Anterior and posterior collecting tubules join bladder arms on a level with middle of ventral sucker. Bladder arms with two loops, each loop with a ciliated patch, close to junction with collecting tubules.

Host: Gyraulus parvus (Say).

Locality: Sun Prairie, Dane County, Wisconsin, U. S. A.

Cercaria gyrauli possesses in common with *C. elvae, C. physellae, C. stagnicolae* and *C. ocellata* the same number of penetration glands, an identical excretory system, and dorso-ventral furcal fin folds. It can be distinguished from these by the shorter but wider body, the shorter and narrower tail stem, and the shorter furcae. In a general way *C. gyrauli* resembles the cercaria of *B. polonica* very closely but the latter has five rather than six pairs of penetration glands and its body is larger. No confusion exists between *C. gyrauli* with six pairs of flame cells in the body and the cercaria of *B. yokogawai* with seven pairs or with *C. tuckerensis* and the cercaria of *S. douthitti* with five pairs. Likewise, because of the difference in excretory systems and general body proportions there is no confusion between *C. gyrauli* and *C. elongata.*

In its activity *C. gyrauli* would be confused only with the cercaria of *B. polonica* or the cercaria of *S. douthitti* because, like them, it clings to the under side of the surface film of the water.

As in *C. elongata* the ventral sucker of this cercaria is relatively small and inconspicuous but it is situated more posteriorly. The penetration glands usually stain well with a combination of neutral red and Nile blue sulphate. The posterior three pairs are more heavily granular than the anterior two pairs. No escape glands were seen but the specimens dissected from snail tissue to check this point were not in good condition. The cells of the genital primordium are conspicuous with large clear

[3] Except where indicated measurements are of 32 specimens killed in four groups (5, 15, 8 and 4) and the character of the variation expressed in the same manner as for *C. elongata.*

nuclei and prominent nucleoli. The muscle strands in the tail stem are prominent and there is no suggestion of caudal bodies as was mentioned in the description of *C. elongata.* The sporocysts were not studied in detail but no structures out of the ordinary were noted.

DISCUSSION

The incidence of these two schistosome cercariae cannot be expressed in percentages because the collections did not represent the entire range in size and age of the snail host. After preliminary examinations which indicated that only the larger of the snails harbored these cercariae only the larger snails were taken in the collections. Of the 373 *G. parvus* from the Sun Prairie pond that were examined carefully 58 were infected with *C. gyrauli* and 26 with *C. elongata.* In addition there were 7 immature infections which were probably schistosomes since other trematodes were extremely rare in this group of snails. At no time was it possible to find enough specimens of this snail in the Sun Prairie pond to make examinations that were completely adequate for an analysis of the seasonal incidence or variation of the infections.

A special effort was made to collect *G. parvus* and related planorbids from other localities in Wisconsin. In all, 1449 individuals of a number of species of the smaller planorbids, of which 439 were *G. parvus,* were examined from fifteen localities and none were found to harbor either of the two species of schistosomes here being considered. These collections were made from areas that were reasonably similar to the Sun Prairie pond; i.e., marshes and marshy lakes. No explanation for this apparently restricted distribution of these two cercariae appeared.

Several attempts were made to determine if either of these two cercariae produce a dermatitis in man. The human host used was known to be susceptible to the development of a dermatitis following penetration of other non-human schistosome cercariae. Only a single lesion was obtained from several mild exposures to *C. elongata* while no lesions resulted from several exposures to *C. gyrauli.* The experiments with the latter cercaria are not completely conclusive since this species attaches so readily and firmly to objects that it is difficult to transfer them to the host's skin. Further evidence of dermatitis produced by these cercariae was obtained during the collections made later in the summer at Sun Prairie pond. At this time no other dermatitis-producing cercariae could be found yet a number of typical lesions arose on those parts of the collector's skin that had been in the water. It would be impossible from this evidence to determine if both of the two species of cercariae emerging from *G. parvus* are capable of producing dermatitis. However, it is certainly safe to conclude that snails of the species *G. parvus* might become involved in outbreaks of "swimmer's-itch" and that they will have to be considered in subsequent studies of this disease.

An effort is being made to determine the life cycles of these two species of schistosome cercariae.

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A "TICKTORIUM" FOR THE PROPAGATION OF A COLONY OF INFECTED *ORNITHODOROS TURICATA*

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A feasible method of maintaining a laboratory colony of ticks of the genus *Ornithodoros* is of value not only for a study of the biology of the tick, but also as a means of maintaining the relapsing fever spirochetes for which the tick serves as a vector, and which are difficult if not impossible to cultivate in vitro. Since the spirochete harbored by the ticks are highly infectious for man the element of safety in the maintenance of the colony is an important factor.

Weller and Graham (1930) and Graham (1931) established the vector relationship of the tick *Ornithodoros turicata* Dugès to human cases of relapsing fever in central Texas. The infected ticks were found in a shallow cavern near the mouth of Falls Creek in the southeast corner of San Saba County, Fig. 1. For a number of years this cave served as a convenient source of infected ticks and many workers have used the ticks and/or spirochetes originating from this source in their published experiments—Weller and Graham (1930), Graham (1931), Bohls and Schuhardt (1933), Kemp, Moursund and Wright (1933) (1934a) (1934b), Brumpt (1933), and Francis (1938). This recent paper by Francis includes a description of his methods of propagation and handling of infected ticks, and points out the important fact that both the ticks and the spirochetes they harbor are exceptionally long lived.

In 1936 it became obvious that the waters of the newly created Buchanan Lake on the Colorado River would inundate the tick-infested cave near Falls Creek. Consequently a final collecting trip was made to the cave in October of 1936, at which time several hundred ticks were collected. At the same time about two gallons of the powdery limestone dust into which the ticks burrow was obtained from the floor of the cavern. It then seemed advisable to attempt to build a safe and suitable housing device which would approximate the natural habitat of the ticks, thereby permitting certain ecological studies and at the same time maintaining the colony with a minimum of effort and danger. This housing device (which I choose to call a "ticktorium") has been in operation now for three years, and for this period has proved entirely satisfactory.

CONSTRUCTION OF TICKTORIUM

(See Figs. 2, 3, 4)

The ticktorium is essentially a miniature cavern in a $14 \times 12 \times 9\frac{1}{2}$-inch block of Burnet County limestone. The limestone block rests on a two-

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inch-thick board which covers a 1½-inch layer of sand in the bottom of a galvanized iron box housing the structure. A two-inch-wide tongue across the rear of the limestone block extends down to and makes contact with the sand. The floor of the cavern and the area in front and to the sides of the limestone block is covered with a two- or three-inch layer of the limestone dust from the cave near Falls Creek. This limestone dust is held in place by a two-inch-wide strip nailed to the floor board about 3 inches in front of the limestone block. A bung hole bored through the floor board in front of the dust-retaining strip permits the pouring of water into the sand layer below the floor. It was assumed that the water would pass from the sand into the tongue of the limestone block and thereby aid in maintaining a suitable humidity in the cavern.

The galvanized iron box housing this miniature cavern is constructed of heavy gauge metal, 24 inches long by 15 inches wide and 13 inches deep. It has a recessed trough one-half inch deep by three-fourth inch wide around the inside near the top. This trough is partially filled with a vaseline-oil mixture and serves as a primary trap to catch ticks attempting to escape from the ticktorium. The box is covered with a 1-inch angle-iron lid ($16\frac{1}{4} \times 25\frac{1}{4}$ inches outside dimensions) which has a layer of ¼-inch mesh hardware cloth and an additional layer of 16-to-the-inch screen wire soldered in place on top of the angle iron. The screen wire is an important precaution against the entrance of flying insects which might easily carry the tiny larvae and nymphs from the ticktorium. The housing box is set in a shallow galvanized iron pan ($27 \times 18 \times 1\frac{1}{2}$ inches) which is partially filled with crank case oil and is designed to serve as a secondary trap should a tick ever get by the primary trap at the top of the box.

MAINTENANCE OF TICKTORIUM

Unfortunately no exact count as to number or sex was made on the initial tick plant. Roughly about a hundred nymphs and adult ticks were placed in the ticktorium on November 20, 1936. There is no way of knowing the exact extent of the present tick population, but it is well up in the thousands. Additional thousands of larvae and nymphs and a few adults have been trapped in the oil of the primary trap. There is no evidence that a tick has ever gotten past this trap.

Francis (1938) has pointed out the extreme resistance of *O. turicata* to starvation. No such rigorous starvation periods have been imposed on the ticktorium colony. However, the feeding intervals (1 ± months to 7 months) have been spaced with regard to convenience rather than the possible needs of the tick colony. Many of these feedings were primarily for the purpose of obtaining spirochete-infected animals for various experiments. White rats have been used exclusively for feeding the ticks. The rat is first placed in a hardware cloth (½-inch mesh) cylinder

which is then stoppered at both ends with No. 10 rubber stoppers. A strand of copper wire ensures the stoppers remaining in place while the rat is in the ticktorium. The caged rat is then placed on the dirt in front of the cavern and is left there for varying periods of time. Table 1 indi-

TABLE 1.—*Tick feedings*

Date of feedings	Duration of exposure of rats to ticks	Effect on rat	Infectivity of tick bites
Nov., 1936	Rat in wire basket overnight	Lived	Infected
Dec., 1936	" " " " "	"	"
Jan., 1937	Rat in cylinder* "	"	Not examined
Feb., 1937	" " " "	"	" "
Mar. 6, 1937	" " " "	"	Inf. 3–10–37
Mar. 23, 1937	" loose in Ticktorium "	"	Not examined
Apr. 3, 1937	Rat in cylinder "	"	Not examined
Apr. 21, 1937	" " " "	"	Inf. 4–24–37
May 6, 1937	" " " "	"	Inf. 5–11–37
May 29, 1937	" " " "	"	Not examined
July 13, 1937	" " " "	Died	.....
Aug. 22, 1937	" " " "	Lived	Not examined
Oct. 12, 1937	" " " "	Died	.....
Oct. 13, 1937	" " " "	Lived	Not examined
May 26, 1938	" " " "	Died	.....
Oct. 14, 1938	" " " "	Died	.....
Oct. 24, 1938	" " " 5 hours	Lived	Inf. 10–28–38
Dec. 14, 1938	" " " 2 "	"	Inf. 12–17–38
Feb. 20, 1939	" " " 3¼ "	"	Inf. 2–24–39**
Apr. 28, 1939	" " " 1 hour	"	Inf. 5– 1–39
June 13, 1939	" " " ¾ "	"	Inf. 6–16–39
June 13, 1939	" " " 2 hours	"	Inf. 6–17–39
Sept. 12, 1939	" " " overnight	"	Inf. 9–17–39
Oct. 6, 1939	" " " 1 hour	"	Inf. 10– 9–39
Nov. 1, 1939	" " " ½ "	"	Inf. 11– 6–39
Nov. 13, 1939	" " " < 4 hours	Died	.....

* Hardware cloth cylinder stoppered at both ends.
** Rat found to be previously infected with *Trypanosoma lewisi*.

cates the dates and duration of the individual feedings and the subsequent effect on the rats. It is interesting to note that by July 1937 the tick population had so increased that an overnight stay in the ticktorium was fatal to the rat. At the most recent feeding (November 13, 1939) the rat was dead in less than four hours after exposure to the ticks.

Removing the rats from the ticktorium demands the greatest precaution to prevent the escape of ticks. The majority of the ticks will feed and detach from the animal within 15 minutes to 1 hour after attaching. However, there are always late comers still in the act of feeding, and a small percentage of the ticks remain attached for varying periods up to several days. Consequently deticking is necessary. For this purpose after the desired feeding interval the hardware cloth cylinder containing the rat is suspended free in the ticktorium by means of two strands of wire with small hooks at each end. The upper hooks are attached to a cross bar which rests on the inner sides of the primary oil trap. The rat is left thus suspended in the ticktorium for several hours to overnight, during which time most of the ticks complete their feed and fall to the floor of the ticktorium.

As a further precaution the live rats, after the deticking period, are transferred to a tall glass cylinder (18 inches high × 11½ inches diameter)

having a hardware cloth false bottom and a hardware cloth lid (see Fig. 3b). The top inside of the glass cylinder is smeared with a vaseline-oil mixture and the cylinder is set on a pan with an oil trap surrounding it. The ticks that leave the rat after removal to the glass cylinder (mainly larvae and young nymphs) are either trapped in the excreta in the bottom of the cylinder where they are later killed, or they can be collected from the inner surface of the glass as they attempt to climb the walls of the cylinder. Careful examination of rats at intervals in the glass cylinder have never revealed ticks remaining attached longer than three days. The rats, however, are kept in the glass cylinder for a minimum of 10 days and then are dipped in an oil bath as an added precaution. Rats killed by the ticks during feeding are transferred to a pail with a tight fitting lid and autoclaved. An especially designed galvanized iron pan (22 inches × 28 inches) which is surrounded by an oil-filled trough aids in safely transferring rats from the ticktorium to the glass cylinder or pail (see Fig. 3c). It also makes a safe working surface for the collection of blood from the exposed rats and for a variety of experiments involving the feeding or handling of infected ticks.

The fact that both temperature and humidity affect the life habits and longevity of the Ixodoidea is common knowledge and it is possible that the temperature-humidity requirements of different species of the genus *Ornithodoros* will be found to vary. The ticktorium colony of *O. turicata* has been maintained at room temperature on a table in my office. This temperature, due to steam heat in the winter, has probably never dropped below 60° F, whereas in summer the temperature has ranged to a probable maximum of 102° F. As previously stated an effort has been made to maintain an increased humidity within the ticktorium by the addition of water to the layer of sand in the bottom. The intervals between these additions have been very irregular (once every month or so) and no check has as yet been made on the variation in humidity. The results, however, warrant the conclusion that the humidity is adequate for *O. turicata*.

SUMMARY

Details of construction and maintenance of a "ticktorium" for the propagation of a spirochete-infected colony of *Ornithodoros turicata* are given. Also methods for exposing animals to infection and for handling them after infection are explained.

This ticktorium has made a constant source of ticks and relapsing fever spirochetes easily available for experimental purposes during the past three years.

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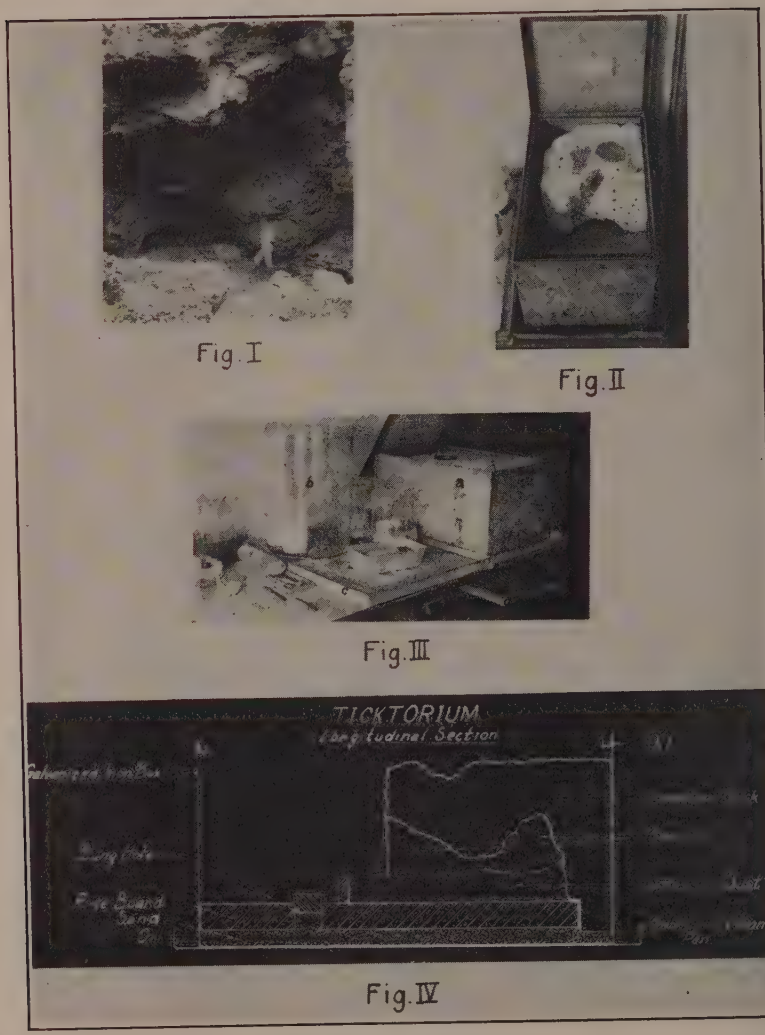

EXPLANATION OF PLATE

FIG. I. Entrance to Falls Creek Cave.
FIG. II. Ticktorium.
FIG. III. Accessory paraphernalia.
FIG. IV. Diagram of ticktorium.

STUDIES ON STRONGYLOIDES. VI. COMPARISON OF TWO HOMOGONIC LINES OF SINGLY ESTABLISHED *S. RATTI*

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In *Strongyloides ratti*, a free-living bisexual generation may alternate with a parasitic generation composed only of females. Such a process has come to be called the heterogonic, or indirect, life cycle. On the other hand, a homogonic, or direct, life cycle may occur in which the progeny of the parasitic generation metamorphose directly into infective filariform larvae. The latter are then ready to infect a new rat host. These direct development larvae are morphologically indistinguishable from those produced by the free-living, bisexual generation.

There is a fundamental question involved in the life cycle of this parasite—and it applies to other species in the genus as well: Is the direction of development in the progeny of the parasite determined at the time the egg is formed or is it determined at a later time, i.e., is the destiny of the young larvae determined by the environment in which it develops? Beach (1936) contended that the mode of development of *S. simiae* larvae was not predetermined in the egg, but was governed by "influences" in their developmental environment. Faust (1936) considered it "significant" that this reported behavior on the part of *S. simiae* was "apparently not based on genetic factors," being contingent instead solely "on environmental factors."

With the demonstration by Graham (1935, 1936) that *S. ratti* could not only be established as a fertile parasite by exposing a rat to a single homogonic larva, but also maintained at this minimal level by serial passage, a new technical method for studying the bionomics of this parasite became available. On the basis of these studies it was established that in all phases the life cycle of *S. ratti* could be encompassed by the reproductive performance of a *single* parasite. Continued study of the daily reproductive activity of these singly established parasites produced evidence which made it difficult to believe that determination of the mode of larval development was accomplished by influences acting on the larvae during their period of free-living development (Graham, 1938a).

When Graham (1939b) compared a homogonically derived line of singly established *Strongyloides ratti* with a similarly established, heterogonically derived line and pointed out biological characteristics which conclusively demonstrated the constitutional dissimilarity of these two

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lines, the idea of "environmental influence" as a critical element in determining the mode of larval development in this species become too heavily burdened with disharmonious facts to merit further serious consideration. While nothing is specifically known concerning possible "genetic factors," it is assumed that they may be involved in the observed "constitutional dissimilarity." Recognition of certain marked differences in the relative proportions of direct and indirect progeny between these two lines occurred with the establishment of the first *S. ratti* of heterogonic origin in a rat host, and continued serial passage brought other differences to light. This fact prompted an investigation of the potentialities of *homogonic* larvae derived from the initial parasitic generation of this *heterogonically* derived line.

In discussing the progeny relationships of a homogonically derived line of singly established *S. ratti,* Graham (1938a) stated that "evidence from the study of additional single larva infections of *S. ratti* of homogonic origin suggests that all homogonic larvae are not constitutionally equivalent." The factual basis for the above statement has since undergone considerable expansion, and data bearing on the point are herein presented in detail.

MATERIALS AND METHODS

One homogonic line of singly established *S. ratti* has been studied for several years (Graham, 1935, 1936). In subsequent sections of this paper this line will be referred to as "direct line I." The new homogonic line, derived from the first parasitic generation of a heterogonically derived line and serially passed and studied in parallel with that line, will be referred to as "direct line II."

Descriptions of technical procedures with the homogonic line (direct line I) have been previously reported (Graham, 1936, 1938a and b, 1939a). Techniques used with the heterogonic line were presented in Graham, 1939b.

With the new homogonic line (direct line II) a combination of previous technical practices and precautions has been followed. The 24-hour fecal collections (daily cultures) were isolated after an incubation period of 24 hours, as were the cultures from the heterogonic line. This was necessary because of the heavy yields of free-living adults. Thus no confusion in enumerating and classifying the progeny as to mode of development occurred. With incubation periods of 48 and 72 hours reliably differentiated counts of the progeny were difficult to obtain due to the rapid development of filariform larval progeny of the free-living bisexual adults.

To obtain infective filariform larvae for the exposure of rats to a single homogonic larva each, cultures were isolated after an incubation period of 24 hours and the recovered progeny permitted to stand in tap

water in 50 cc centrifuge tubes for an additional 24 or 48 hours. This procedure effectively inhibited the development of indirect larval progeny of the free-living adults while giving additional time to the larvae of direct development for their last ecdysis. This had seldom occurred after an incubation period of only 24 hours. As a further precaution against the use of non-infective larvae for the exposure of new rats, each larva used for infection purposes was examined carefully to ascertain that it had exsheathed.

The same strain of inbred laboratory rats was used as reported for previous studies in this series. The conditions under which those harboring single *S. ratti* of direct line II were cared for were the same as described by Graham (1939b) for the parallel heterogonic line as well as for that portion of the infections in direct line I with which calendar comparability was effected.

RESULTS

The characteristics of the singly established *S. ratti* in direct line II so closely resembled those of *S. ratti* in the parent heterogonic line from which this homogonic line was derived that it was impossible to distinguish between them on the basis of single infections. This was readily possible in the case of single *S. ratti* of the homogonic line, i.e., direct line I, and the heterogonic line (Graham, 1939b, Fig. 1). However, there was one point of difference which was of a magnitude and constancy permitting differentiation of the two series of infections, thus indicating that new characteristics may be revealed through the agency of heterogony in *S. ratti.*

In view of the several points of similarity between *S. ratti* of direct line II and the heterogonic line, marked dissimilarity of the two homogonic lines, i.e., direct lines I and II, was naturally evident. The points of difference between these two lines are the same as those enumerated by Graham (1939b) for *S. ratti* in direct line I and the heterogonic line, namely: (1) the relative yields of direct and indirect progeny, (2) the reproductive rate (fecundity), (3) the length of reproductive life, (4) the frequency of production of free-living males of the bisexual generation, and (5) the infection rate.

Since repeated reference to the points of difference in the characteristics exhibited by *S. ratti* from a homogonic line (direct line I) and a heterogonic line (Graham, 1939b) has been and will be made, it seems desirable to provide a brief catalog of major items of difference. This is given in Table 1 which incorporates the data presented in Graham (1939b, Table 1) together with the additional data which accumulated for these two lines from July 1, 1938, until March 7, 1939, when study of the heterogonic line was discontinued. Changes between Table 1 (Graham, 1939b) and Table 1 of this study are thus quantitative in na-

TABLE 1.—*Comparison of progeny relationships and other points of difference in a homogonic line (direct line I) and a heterogonic line of* Strongyloides ratti *from September 22, 1937, to March 7, 1939*

	Direct line I	Heterogonic line
No. of parasites	34	25
Serial generations	20–57	1–12
No. of cultures with one or more offspring	2,559	1,229
Progeny		
Direct larvae	16,502	2,530
Indirect males	10	6,102
Indirect females	3,288	16,300
Total progeny	19,800	24,932
Percentage of indirect progeny	16.7%	89.9%
Ratio (approximate) of males to females	1 : 300	1 : 3

ture and serve to emphasize the fact that the biological differences between these two lines of *S. ratti* are constant.

1. *Differences in the Relative Yields of Direct and Indirect Progeny*

The relative yields of the two progeny types, i.e., larvae of direct development and bisexual adults of the free-living generation, in the two lines under comparison (direct lines I and II) were markedly disproportionate. Evidence of this is shown in Table 2 in which it may be seen

TABLE 2.—*Comparison of progeny relationships and other points of difference in two homogonic lines (direct lines I and II) of* Strongyloides ratti *from October 12, 1937, to September 10, 1938*

	Direct line I	Direct line II
No. of parasites	25	26
Serial generations	31–47	1–13
No. of cultures with one or more offspring	1,666	1,798
Progeny		
Direct larvae	9,057	2,900
Indirect males	9	7,567
Indirect females	1,767	23,187
Total progeny	10,833	33,654
Percentage of indirect progeny	16.4%	91.4%
Ratio (approximate) of males to females	1 : 200	1 : 3

that only 16 per cent of the progeny secured from single *S. ratti* of direct line I were adults of indirect development, whereas 91 per cent of those obtained from the parasites of direct line II were indirect forms. It is to be noted that precise calendar comparability existed.

The average daily yield of each type of progeny calculated by weeks of parasitic age for 26 *S. ratti* in direct line II is shown in Fig. 1. In essential respects the curves almost exactly reduplicate similar curves for *S. ratti* of the parent heterogonic line (Graham, 1939b, Fig. 2). The impossibility of differentiating between these two lines of *S. ratti* on the basis of progeny type relationships alone is obvious. On the other hand comparison of the curves of Fig. 1 with similarly derived curves for *S.*

FIG. 1. Average number of free-living adults and larvae of direct development from 26 single *S. ratti* parasites in direct line II, as observed in cultures made from 24-hour fecal collections from the rat host.

ratti in direct line I (Graham, 1938a, Fig. 2, p. 228) shows the similarity of the curves of the principal progeny type but with the important distinction that in direct line I this type is of direct development whereas in direct line II it is of indirect development.

2. *Differences in the Reproductive Rate (Fecundity)*

It is evident from inspection of the curves mentioned in the preceding section that the reproductive rate of the *S. ratti* in direct line II and the parent heterogonic line must be very similar and that the rate of reproduction in these lines must differ somewhat from that of the *S. ratti* in direct line I. Graphic presentation, however, makes these points clearer. The cumulative progeny yields of Fig. 2 are derived (1) from data on

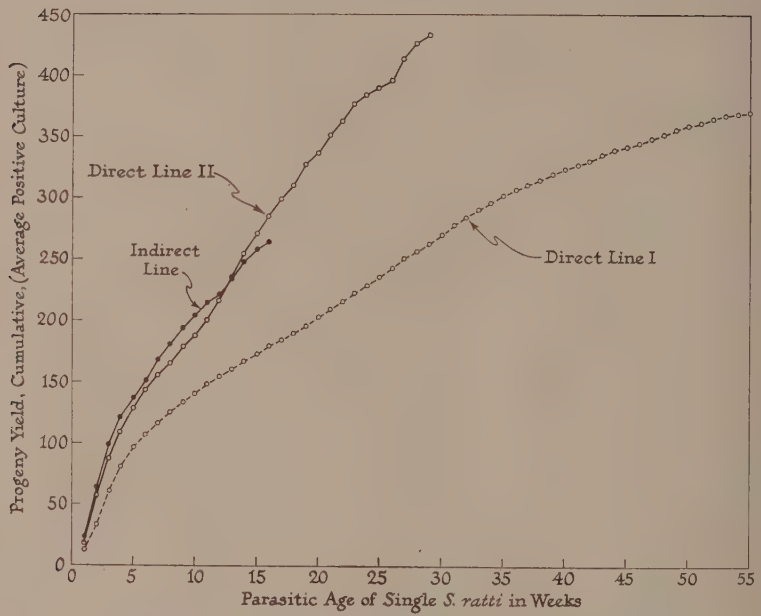

FIG. 2. The cumulative progeny yield from 26 *S. ratti* in direct line II, 116 *S. ratti* in direct line I, and 25 *S. ratti* in the heterogonic line (from which direct line II was derived). Weekly accumulations are comparable, being based on the average yields of positive cultures. In direct line II, 1,798 culture days yielded 33,654 progeny; in direct line I, 5,118 culture days yielded 54,867 progeny; in the heterogonic line, 1,229 culture days yielded 24,932 progeny.

direct line II as given in Fig. 1, (2) from data on direct line I (Graham, 1938a, Fig. 2), and (3) from information on the heterogonic line (Graham, 1939b, Fig. 2). Also included with the latter are data on the heterogonic line from July 1, 1938, to March 7, 1939. In each instance the weekly increment is based on the average progeny output from patent culture days to provide comparability.

The parallelism of the reproductive rate in the heterogonic line and in direct line II is clear as is the lower rate of reproduction which occurred in direct line I. At the end of 16 weeks (indicated in Fig. 2 by arrow) direct line II reached a cumulative progeny level not attained by direct line I until 16 weeks later (indicated in Fig. 2 by arrow).

3. *Differences in the Length of Reproductive Life*

The length of reproductive life of singly established *S. ratti* of direct line I has been presented in detail (Graham, 1940). For 78 infections with good end-points, i.e., 30 or more consecutive culture days without progeny after the last patent day of each infection, a mean reproductive

life span of 149 ± 8 days was observed. In the heterogonic line, the length of reproductive life of *S. ratti* was distinctly less than this, being 11 weeks (Graham, 1939b). Actually, 17 infections with good end-points showed a mean life span of 72 ± 4 days. The difference in the length of reproductive life in these two lines is significant statistically.

In direct line II, as may be observed in Fig. 2, the *S. ratti* were reproductively longer lived than those in the heterogonic line but shorter lived than those in direct line I. Twenty-two infections for which good end-points of reproductive activity were obtained showed a mean life span of 90 ± 8 days. The differences in the mean reproductive life spans of *S. ratti* in direct lines I and II are statistically significant, but the differences between the heterogonic line and direct line II are insignificant. It is possible of course that this may be accounted for by the small number of infections in these two lines. Regardless of mathematical considerations, the fact remains that several of the *S. ratti* in direct line II were reproductively active for a considerably longer period (the longest life span in the group being 199 days) than any in the parent heterogonic line. Prolonged observation of additional parasites in these lines might show valid statistical differences in the length of their reproductive lives.

4. *Differences in the Frequency of Production of Males of the Free-living Bisexual Generation*

As in the heterogonic line, the rate of production of free-living males in direct line II was high, being 22.5 per cent of the total progeny obtained from this line. In the heterogonic line 24.5 per cent of the total progeny were free-living males. On the other hand, the frequency with which free-living males were observed in direct line I was almost negligible, being less than 0.1 per cent of the total progeny observed.

Attention is directed to the similarity of the ratio of approximately 3 females to 1 male in the bisexual progeny from *S. ratti* in the hetergonic line and direct line II, as shown in Tables 1 and 2. (Actual ratios were 2.7 in the former and 3.1 in the latter.)

5. *Differences in Rate of Infection*

An infection rate of 26.2 per cent was reported from the exposure of 668 rats to a single homogonic larva each in direct line I (Graham, 1939b). During more than 4 years since the first infections in this line were initiated, the cumulative infection rate has varied but slightly. A similar constancy has likewise been noted in the cumulative infection rates in the heterogonic line and direct line II ever since these two lines were established. In the heterogonic line, which was passed through 12 serial generations, 64 rats were exposed to a single larva each and 25 infections (39 per cent) obtained. In direct line II, derived from the heterogonic line, the exposure of 35 rats to a single homogonic larva each during

passage of the line through 13 serial generations yielded 26 (74 per cent) positive infections. The writer's confidence in the high infection rate in this line became so great that in a number of instances only a single rat was exposed to a larva for establishing a new generation, and only once was failure encountered in doing so.

As pointed out earlier (Graham, 1939b), the difference in the infection rates in the heterogonic line and direct line I (39 and 26 per cent, respectively) was not sufficiently great and the number of exposures in the heterogonic line was too few to permit of statistical validity even if the difference was actually real. However, another situation prevails in the case of the high infection rate obtained in direct line II even though the number of exposures was small. Here it can definitely be stated that there is only one chance in 200 that the true infection rate might be lower than 50 per cent or higher than 90 per cent. In the heterogonic line there is only one chance in 200 that the true infection rate might be lower than 25 per cent or higher than 55 per cent. While there is a slight overlapping of these limits, the statistical odds definitely favor the interpretation that the dissimilar infection rate in the *S. ratti* of these two closely related lines is real rather than apparent.

Although these two lines of singly established *S. ratti* may be so similar in other characteristics (with the possible exception of length of reproductive life) that they are indistinguishable, on this one point, namely, in rate of infection, they appear to be consistently, as well as statistically, dissimilar. The dissimilarity seems accountable only on the basis of constitution.

The difference in the infection rates in direct lines I and II is statistically valid and serves to re-emphasize the now well established fact that "all homogonic larvae are not constitutionally equivalent," as noted by Graham (1938a).

DISCUSSION

The points of difference between the two homogonic lines of singly established *S. ratti* reported herein were clear cut and demonstrated that homogonic larvae of *S. ratti* may differ constitutionally. Far more significant is the fact that these lines, once established, are maintained without apparent change by continued homogonic passage. The seasonal variation in the frequency with which culture days yielded adults of indirect development in direct line I (Graham, 1939a) is not comparable in degree to the progeny type characteristics herein accepted as criteria for differentiating these distinctive lines of *S. ratti*. Constancy of distinguishing characteristics was likewise observed in another homogonic line of *S. ratti*, which was derived from the same heterogonic line of *S. ratti* as direct line II. The details of this additional line will be reported in another paper.

Although the evidence is limited in that only a few lines have so far been demonstrated and studied, the fact that any distinctive lines of homogonically derived *S. ratti* can be recognized makes it seem odd that these lines arise only by way of heterogony and never from a parasitic female by way of homogony. Their constitutional dissimilarity seems clearly established. If, as Sandground (1926) claimed, *Strongyloides ratti* actually is a syngonic organism, and possesses, as it would appear, a variety of segregable characteristics, one may ask the question: why do these pure homogonic lines exhibit such constancy in their characteristics and why do not new lines exhibiting distinctive characteristics arise from them? Inferentially, one possible explanation is that "homogony" is essentially a vegetative mode of propagation achieved without fertilization of the ova. The observation of Chitwood and Graham (1940) to the effect that parasitic *S. ratti* are parthenogenetic forms seems significant in this connection.

The contrast between the results of the present study and the mass selection experiments of Sandground (1926) with *S. ratti* is extreme. In passage through 10 serial indirect generations Sandground observed but one instance in which free-living adults exceeded larvae of direct development among the progeny. In 12 mass infections in a direct development series of 11 serial generations none produced more indirect than direct progeny, thus being somewhat comparable to direct line I of this study. Sandground observed: "If the original infection, displaying both modes of reproduction with a preponderance of direct development, be regarded as a type characteristic, it is seen that this characteristic is conserved or transmitted through the medium of larvae of direct development." Insofar as his experiments with *S. ratti* are concerned this principle seems to apply almost equally well to larvae of indirect development.

With *Strongyloides papillosus* in the rabbit, Sandground (1926) observed preponderantly direct development in infections established with larvae of direct development obtained from a sheep infection. When selection for indirect development from these homogonically established infections was made, infections were obtained which showed preponderantly indirct development. After passage of this heterogonic line through several generations, an infection established in a rabbit with homogonic larvae derived from a parental generation established heterogonically showed predominantly indirect development. Concerning this, Sandground observed that the result was "rather unexpected" although "consistent with the principal enunciated earlier" on the basis of his *S. ratti* selection experiments. Thus with regard to mode of larval development the relationship existing between direct line II and the parent heterogonic line bears a resemblance, at least superficially to that observed by Sand-

ground in the case of *S. papillosus* in rabbits. Closer comparison is unwarranted.

It would be interesting to know the extent to which homogonic larvae from a heterogonically established mass infection which could develop into parasites capable of producing predominantly progeny of direct development might be masked by more highly infective larvae with different potentialities concerning the mode of larval development. It seems possible that some generalizations concerning *Strongyloides* are not safely based on mass infection experiments.

Comparatively little is known concerning the extent to which varieties or strains of nematode species occur in nature. What meager evidence is available bears in part on strain differences as measured in two host species, rather than in a single one. Some plant parasitic nemas may become so well adapted to new host plants that it is with difficulty that they can become re-adapted to a previously favorable host species. Among nematodes parasitic in vertebrates only a few instances of variation in physiological characteristics have been reported. Scott (1929, 1930) demonstrated the existence of two strains of the hookworm, *Ancylostoma caninum,* which showed a high degree of specificity for the dog and cat respectively. On the basis of egg-worm ratios Graham and Porter (1934) showed that a strain of *Nippostrongylus muris* from Texas consistently produced fewer eggs per female than two strains of the parasite recovered from wild rats at Baltimore.

Chandler (1932) suggested "that a parasite may become better adapted to a host other than the normal one," believing that in the course of repeated passage through white laboratory rats *N. muris* became better adapted to the latter than to their normal wild rat hosts. In the experience of Graham and Porter (unpublished data) laboratory rats were found to be at least as favorable hosts for *N. muris* as wild rats. Taylor (1928) found that when the gapeworm *Syngamus trachea,* derived from starlings, was passed through one generation in chickens, the infectivity of the parasite for the latter host was increased, although later Clapham (1935) readily infected chickens with gapeworms of starling origin. Thus the evidence for adaptation among animal parasites is slight, although it seems probable that any change in host strains or host species should lead automatically to selection of characteristics in the parasite both favorable and unfavorable for survival. It seems unlikely that such changes in the constitution of parasites in mass infections would be detected readily unless the changes were of considerable magnitude.

In the case of *Strongyloides ratti* the differences which have been noted were obtained under extremely favorable conditions in a single host species which was a highly inbred and therefore a relatively homo-

geneous line. These differences were thus directly measurable and referable to the constitution of the parasite.

SUMMARY

1. A homogonic line of singly established *S. ratti* (direct line II) showed remarkable similarity in most respects to the heterogonic line from which it was derived but was distinguishable from the parent line on the basis of a higher infection rate. Direct line II was compared with another homogonically maintained line of singly established *S. ratti* (direct line I) from which it differed in a number of respects.

2. Direct line II differed from direct line I in producing predominantly progeny of indirect development (91 per cent), in showing a higher reproductive rate, in being shorter lived, in producing many more males of the free-living generation, and in producing filariform larvae which were nearly three times as infective when exposed singly on rats.

3. The characteristic differences between these two homogonic lines of singly established *S. ratti* indicate clearly that all homogonic larvae of *S. ratti* are not constitutionally equivalent.

4. The characteristics of these homogonically maintained lines of *S. ratti* appear not to be changed during the course of serial homogonic passage.

5. On the basis of present evidence it appears that new lines of *S. ratti* arise only through the agency of heterogony and never by homogony.

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SOME HELMINTH PARASITES OF THE PANAMA OTTER

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On August 22, 1939, Dr. A. O. Foster placed at the writer's disposal a vial containing 4,183 trematodes belonging to the family DIPLOSTOMIDAE. In the same vial there were also seven specimens of a very small trematode belonging to the family HETEROPHYIDAE, as well as a few specimens of a nematode, *Strongyloides* sp. The material was collected February 17, 1939, from the intestine of an otter, *Lutra repanda* Goldman, 1914, by Dr. Foster and his associates, at the Gorgas Memorial Laboratory, Panama, R. P. The host from which the specimens were obtained was found dead, the cause of death apparently being the result of the unusually large number of trematodes present. There are no published records of any parasites from this host, although there are three related species of diplostomids reported from a related host (the Brazilian otter), *Lutra brasiliensis* Zimm. After reviewing the descriptions of the three most closely related species, *Diplostomum alarioides* Dubois, 1937, *Alaria clathrata* (Diesing, 1850) LaRue, 1926, and *A. pseudoclathrata* (Krause, 1914) LaRue, 1926, sufficient differences were noted in Foster's material to justify the establishment of a new species.*

Diplostomum fosteri McIntosh, 1939

Description: Body elongate, up to 4.5 mm long, bisegmented; anterior segment with anterior margin trilobed and with lateral and posterior margins folded ventrally; anterior segment shorter than posterior. In a specimen 4.29 mm long, the anterior segment was 1.52 mm and posterior segment 2.77 mm. Oral sucker terminal 120 μ by 110 μ, smaller than pharynx; lateral pseudosuckers well developed, smaller than oral sucker and located at level of junction of oral sucker and pharynx. Prosdetic glands conspicuous, occupying broad areas lateral to acetabulum. Acetabulum about 85 μ by 90 μ, weakly developed, smaller than oral sucker, located between anterior extremity of tribocytic organ and intestinal fork. Tribocytic organ well developed, elliptical, with ventral longitudinal slit-like opening. Prepharynx short; pharynx almost spherical, about 140 μ in diameter; esophagus short; intestinal ceca about 50 μ in diameter, extending almost to level of bursa copulatrix. Testes in about middle third of posterior segment, width and thickness greater than length; each testis divided posteriorly into five lobes, two lobes ventro-lateral, two dorso-lateral, and one dorso-median; the dorso-median lobe shorter and, in case of anterior testis, less distinct than other lobes. Anterior testis slightly asymmetrical, 340 μ long by 480 μ wide by about 450 μ thick; posterior testis 350 μ long by 570 μ wide by about 540 μ thick. Seminal vesicle large, somewhat convoluted, occupying most of the space between posterior testis and copulatory bursa. Ovary ellipsoidal, broader than long, 100 μ long by 340 μ wide by 100 μ thick, pretesticular, near cephalic limit of middle third of body. Oviduct arising from dorsal side of ovary, extending posteriorly along dorsal and left lateral side of anterior testis to join oötype, the latter situated to left of median line between testes. Laurer's canal, short, S-shaped, arising from oviduct a short distance from ovary and opening dorsally a short distance poste-

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* Described briefly in 1939 (J. Parasitol. 25 Suppl: 25).

rior to the ovarian level. Mehlis' gland and vitelline reservoir situated between testes. Vitellaria extending from near level of anterior margin of tribocytic organ to cephalic margin of anterior testis. In the anterior segment of the body the vitellaria penetrate the tribocytic organ and extend into the ventrally-folded lateral and posterior margins; in the posterior segment the vitellaria are confined to lateral margins. Uterus extending anteriorly from its origin to a point near junction of anterior and posterior body segments, then turning ventrally and proceeding posteriorly as a number of loose coils to genital pore, opening into hermaphroditic canal posterior and ventral to male genital pore. Copulatory bursa subterminal with large cavity opening postero-dorsally; hermaphroditic pore opening at margin of postero-ventral wall of bursa. Eggs 95 to 118 μ by 55 to 68 μ; up to as many as 100 present.

Habitat: Intestine of Panama otter, *Lutra repanda* Goldman, 1914.

Distribution: Panama, R. P.

Specimens: U. S. Nat. Mus. Helm. Coll. No. 44106 (type) and 44107 (paratypes).

The new species separates out with *Diplostomum alarioides* in the key given by Dubois (1938, Mem. Soc. Neuchatel Sc. Nat. **6**: 1–535) in his "Monographie des Strigeida (Trematoda)." The two species may be separated anatomically on a number of points, the most outstanding being the level at which the vitellaria terminate posteriorly; in *Diplostomum alarioides* they terminate at the level of the copulatory bursa, while in *D. fosteri* they extend only as far as the level of the cephalic margin of the anterior testis. The oral sucker in the former species is larger than the pharynx, and smaller in the latter.

Diplostomum fosteri shows marked resemblance with two species placed in the subfamily ALARIINAE: *Alaria clathrata* and *Alaria pseudoclathrata*. *A. clathrata* resembles *Diplostomum fosteri* in that the pharynx is larger than the oral sucker, the lateral pseudosuckers are similarly placed, and the two species are about the same in body length. In the case of *Alaria pseudoclathrata* there is still further resemblance to *Diplostomum fosteri* in that the posterior surface of the testes is divided into a number of lobes.

Both *Alaria clathrata* and *A. pseudoclathrata,* as well as *Diplostomum alarioides* and *D. fosteri,* all parasites of otters, have so many points in common that it would appear that all four species should be members of the same genus. Actually, however, the four species, if Dubois' (loc. cit.) classification is to be followed, are split into two subfamilies, namely, ALARIINAE and DIPLOSTOMINAE. Both *Alaria clathrata* and *A. pseudoclathrata* have the posterior body segment shorter than the anterior, and the uterus penetrates the tribocytic organ; these have been placed in the ALARIINAE Hall and Wigdor, 1918. On the other hand *Diplostomum alarioides* and *D. fosteri* have the posterior body segment longer than the anterior, and the uterus does not penetrate the tribocytic organ; these fall into the subfamily DIPLOSTOMINAE Monticelli, 1888.

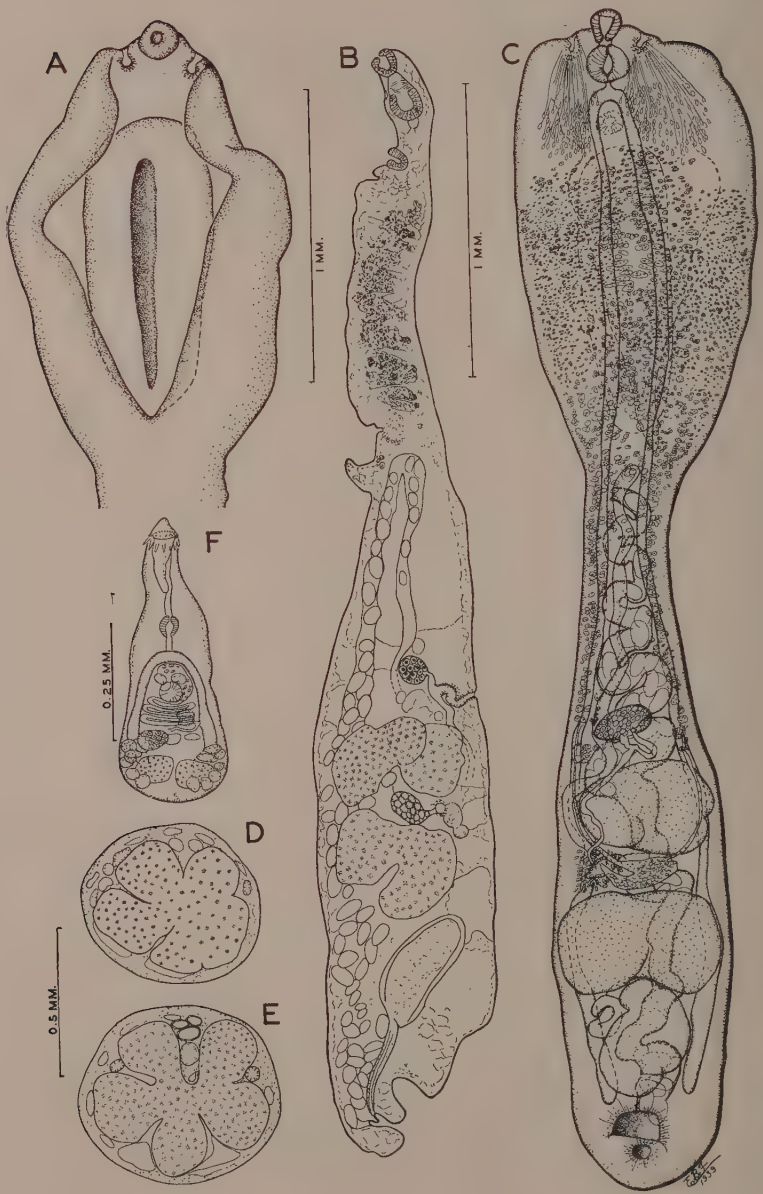
Phagicola longa (Ransom, 1920) Price, 1932

Among the specimens of *Diplostomum fosteri* were found 7 specimens (U. S. Nat. Mus. Helm. Coll. No. 44104) of a very small trematode

belonging to the subfamily CENTROCESTINAE. After comparing these specimens with the type material of *Phagicola longa,* which was from an Alaskan fox, and also with specimens from Palestine dogs, the only difference appears to be that of size. The specimens from the otter apparently represent a diminutive race of *Phagicola longa,* the length being only 450 μ which is about half that of specimens from the fox and the dog.

EXPLANATION OF PLATE, p. 222

A–E: *Diplostomum fosteri* McIntosh, 1939.
A. Outline of anterior segment, ventral aspect.
B. Longitudinal section showing arrangement of reproductive organs.
C. Entire specimen, dorsal aspect.
D. Cross-section through anterior testis.
E. Cross-sestion through posterior testis.
F. *Phagicola longa* (Ransom, 1920) Price, 1932, ventral aspect.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A COMPARATIVE STUDY OF THE EGGS OF VARIOUS SPECIES OF NEMATODES PARASITIC IN DOMESTIC RUMINANTS

D. A. Shorb

U. S. Bureau of Animal Industry

Methods of identifying eggs of genera of nematodes occurring in domestic ruminants have been discussed in a previous paper (Shorb, 1939). For most practical purposes the determination of the genera involved is sufficient, but in certain cases it is desirable to know the species of nematode represented. For example, two species of *Oesophagostomum,* namely, *O. columbianum* and *O. venulosum,* may be present in the same sheep. However, only one, *O. columbianum,* is associated with the formation of nodules and is generally regarded as the more pathogenic. Therefore, when *Oesophagostomum* eggs are found, it is desirable to know which species is represented. Other comparable cases are known, and as our knowledge of ruminant parasites increases, the list of such cases may become augmented.

PROCEDURE

The measurements used in the previous paper (Shorb, 1939) are presented in this paper in the form of charts in order that the difference in size of the eggs of the species studied may be readily visualized. These charts also indicate the frequency of distribution within the size ranges of the eggs. Similar charts were given by Wood (1932), but he made no attempt to indicate the size of each individual egg measured, and consequently the rectangular diagrams presented by that author may be misleading, because species having no eggs similar in size to those of another species are actually represented as having such eggs, and the degree of overlapping is exaggerated. This source of error is eliminated in the present charts and diagrams, as each egg measured is represented on the charts by a characteristic symbol. The position of the symbol indicates that the dimensions of the eggs measured lie between the figures given on the chart.

All eggs were measured under a microscope provided with a 10× eyepiece containing a contrast micrometer disk, and with a 4 mm objective. The distances between the divisions of the eyepiece micrometer were equal to 1.633 microns, but in order to avoid the use of the larger fraction the divisions of the charts equal 1.6 microns with each third division equalling 1.7 microns. The scale of the chart representing the eggs of *Nematodirus* is reduced one-half to conserve space.

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Because of the similarity of appearance of different eggs, an absolutely accurate identification of each egg examined is not always possible. In such cases, however, the host and geographical distribution of the parasite may aid in the identification of the species of worm involved. Fifteen genera of nematodes, the eggs of which occur in the feces, have been reported as parasites of domestic ruminants in the United States. Six of

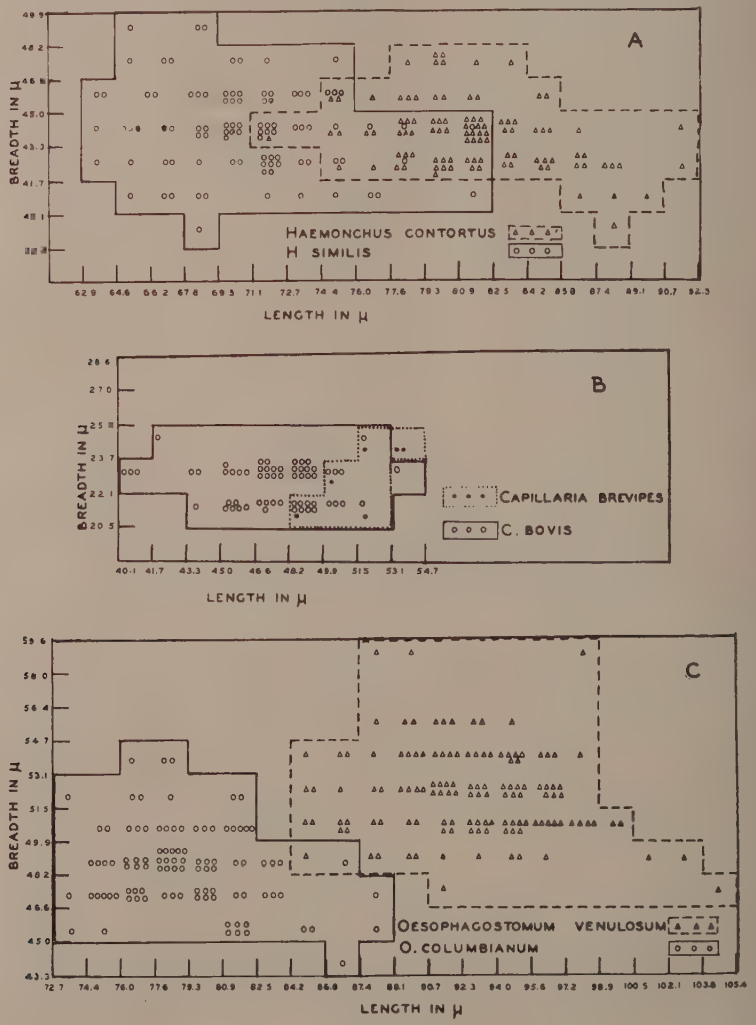

FIG. 1. Distribution of eggs within the indicated size ranges (A) *Haemonchus contortus* and *H. similis;* (B) *Capillaria brevipes* and *C. bovis;* (C) *Oesophagostomum venulosum* and *O. columbianum.*

these, namely, *Chabertia, Neoascaris, Skrjabinema, Strongyloides, Syngamus* and *Trichuris* are each represented by a single species. Since the eggs of these have been previously described (Shorb, 1939) they will not be considered in this paper. Furthermore, since *Bunostomum trigonocephalum* usually occurs in sheep and goats, while *B. phlebotomum* is generally confined to cattle, the eggs of these may be distinguished for the most part on the basis of the host relationship. The other eight genera, namely, *Capillaria, Gongylonema, Haemonchus, Cooperia, Nematodirus, Ostertagia, Trichostrongylus* and *Oesophagostomum,* have more than one species in at least one of the hosts considered, and this paper is restricted to differentiating eggs of species belonging to these genera.

Capillaria spp.

The two species of *Capillaria, C. bovis* and *C. brevipes,* occurring in domestic ruminants in this country have eggs so characteristic that they may be easily distinguished from each other. In the eggs of *C. bovis* the polar plug-like structures project beyond the shell a distance equal to about one-half the length of the plug, while in *C. brevipes* the polar plugs do not project beyond the limit of the shell. The shell of the egg of *C. brevipes* is thicker (3.5 μ) than that of *C. bovis* (2.3 μ). The size distribution of these eggs is shown in Fig. 1, B.

Gongylonema spp.

Two species of *Gongylonema, G. pulchrum* and *G. verrucosum,* are found in cattle, sheep, and goats. The eggs of *G. pulchrum* are so much larger than those of *G. verrucosum* that they may be easily distinguished from the latter species. (Fig. 2, B).

Haemonchus spp.

Two species of *Haemonchus,* namely, *H. contortus* and *H. similis,* occur in domestic ruminants in this country. Although *H. similis* has been reported from sheep in Brazil, it has not been found in this host in the United States, neither are there any records of its occurrence in goats. It does occur to some extent in cattle in Florida, Louisiana, Texas, and Puerto Rico, and in deer in Florida. Therefore, except in these locations and in the surrounding territory, eggs of *Haemonchus* spp. may be considered to be those of *H. contortus.* However, *Haemonchus* eggs from cattle in the Gulf States and Puerto Rico may be either *H. similis* or *H. contortus;* these may be differentiated with the aid of Fig. 1, A. This figure shows the size distribution of *Haemonchus* eggs taken from the uteri of worms from cattle. There is an overlapping of the egg sizes of the two species as shown in the diagrams; this overlapping represents 37 per cent of the eggs measured. However, as no eggs of *H. contortus* are

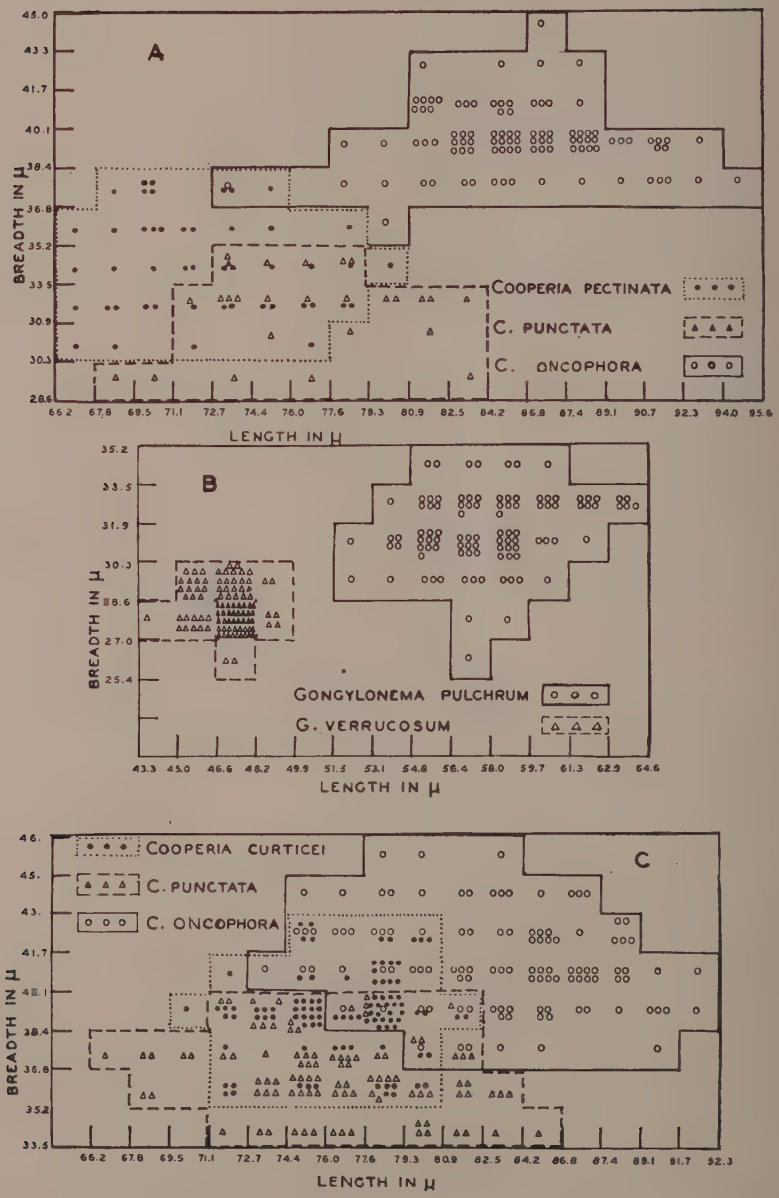

FIG. 2. Distribution of eggs with the indicated size ranges, (A) *Cooperia pectinata, C. punctata* and *C. oncophora;* (B) *Gongylonema pulchrum* and *G. verrucosum;* (C) *Cooperia curticei, C. punctata* and *C. oncophora.*

shorter than 71 μ and none of *H. similis* is longer than 82 μ, it is necessary to measure a number of eggs and compare the distribution with that on the chart in order to determine which species is present. If both species are present, eggs will be found in both size ranges.

Cooperia spp.

Five species of *Cooperia,* namely, *C. curticei, C. punctata, C. pectinata, C. oncophora* and *C. bisonis,* occur in domestic ruminants in the United States. One of them, *C. bisonis* is rare and, therefore, not considered in the present paper. *C. curticei* and *C. oncophora* are common parasites of sheep; the size difference of their eggs are shown in Fig. 2, C. A diagram representing the eggs of *C. punctata* is also included in Fig. 2, C, since this species has been reported from sheep, although it is more commonly found in cattle. Three species of *Cooperia,* namely, *C. punctata, C. pectinata,* and *C. oncophora* have been reported as common parasites of cattle. The problem of differentiating the eggs of these three species is simplified to some extent by the known distribution of *C. pectinata* and *C. oncophora* that of the former being limited to the South and that of the latter to the more northern sections of the United States. When eggs of *Cooperia* appear in the feces of animals from the South, the eggs of *C. pectinata* and *C. punctata* must be considered; in other parts of the United States *Cooperia* eggs are probably those of *C. oncophora* and *C. punctata.* Diagrams indicating the sizes of the eggs of these three species as obtained from the uteri of worms collected from cattle are given in Fig. 2, A.

Nematodirus spp.

Four species of *Nematodirus,* namely, *N. filicollis, N. spathiger, N. abnormalis* and *N. helvetianus* are found in the United States. Two of these, *N. filicollis* and *N. spathiger,* have been reported from cattle, sheep, and goats. The third species, *N. abnormalis* has not been reported from cattle, while the fourth, *N. helvetianus,* has not been reported from sheep and goats. The eggs of *N. spathiger* are not represented in Fig. 3 as they may be easily identified, as pointed out by various authors (Boulenger, 1915; Zawadowsky, 1929; Tetley, 1935), by their thickened, tapering ends. This thickening is due to a deposit of material between two of the layers of the shell. A large proportion of the eggs of *N. helvetianus* have one end of the shell projecting in the form of a low knob, otherwise there appears to be no important morphological difference, except size, between these and the eggs of *N. abnormalis* and *N. filicollis.* As the eggs of *N. spathiger* can be separated from the others by the presence of polar thickenings, and *N. helvetianus* and *N. abnormalis* from each other by host distribution, it is only necessary to separate the last two from *N. filicollis.* This may be accomplished with a fair degree of certainty on the basis of size difference as shown in Fig. 3.

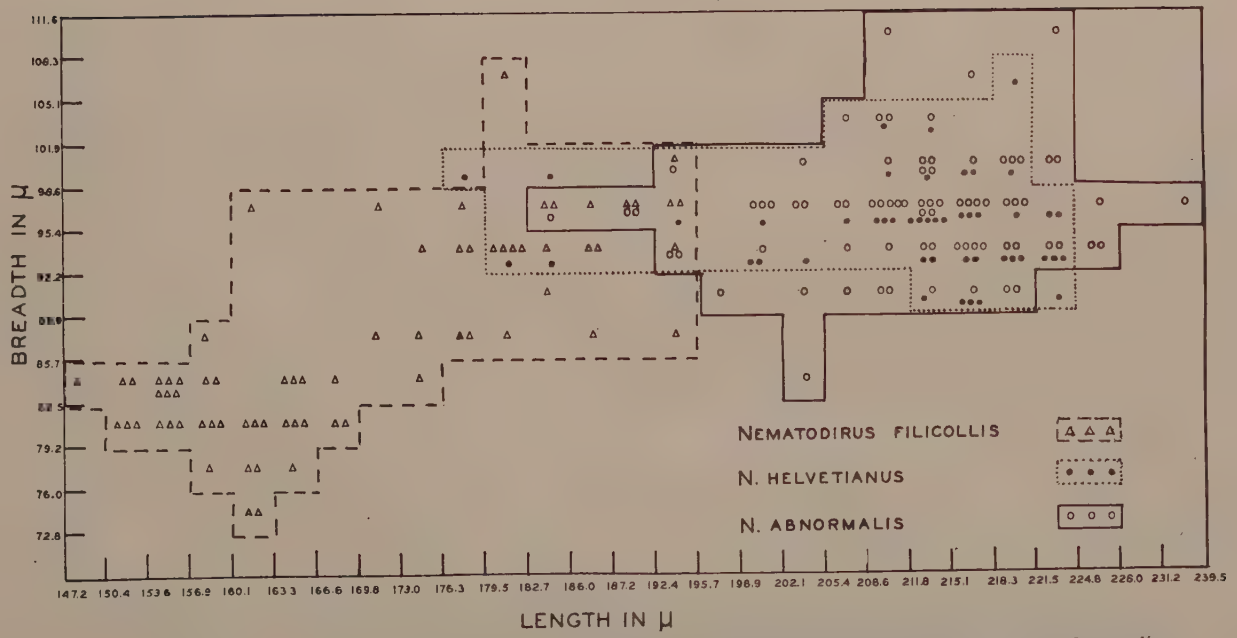

FIG. 3. Distribution of eggs within the indicated size ranges, *Nematodirus filicollis, N. helvetianus* and *N. abnormalis.*

Ostertagia spp.

The females of *Ostertagia* spp. have not been adequately differentiated morphologically and, therefore, collection of eggs from known species is impossible at present. However, the identification of eggs of species of *Ostertagia* in feces may be accomplished to some extent on the basis of host and geographical distribution. Two species, *O. ostertagi* and *O. lyrata* are largely confined to cattle; the former is not confined to any particular part of the United States, whereas the latter is, so far as known, confined to the South and has been reported only from Louisiana and Virginia. Two species, *O. circumcincta* and *O. trifurcata* occur in sheep and goats in all parts of the country, while *O. marshalli* and *O. occidentalis* are found in sheep in the Western States. Moreover, the eggs of *O. marshalli* are so much larger than those of any of the other species of *Ostertagia* that they may be identified on the basis of size alone.

Oesophagostomum spp.

Three species of *Oesophagostomum,* namely, *O. radiatum, O. venulosum,* and *O. columbianum* occur in the United States. *O. radiatum* has not been reported from sheep and goats, while *O. columbianum* and *O. venulosum* are confined to those hosts. Consequently, it is only necessary for the present to differentiate eggs of the last two species. Fortunately, the sizes of only 0.9 per cent of the eggs of these species overlap (Fig. 1, C) and for that reason little difficulty should be experienced in separating them.

Trichostrongylus spp.

Trichostrongylus axei, T. colubriformis, T. vitrinus, T. capricola and *T. longispicularis* occur in domestic ruminants in the United States. One of these, *T. longispicularis,* is rare and no females or eggs were available for this study. Of the others, *T. capricola* has not been reported in cattle and occurs rarely in sheep; the other species have been obtained from cattle, sheep, and goats. There is usually sufficient variation in size and shape of the eggs to enable the species of *Trichostrongylus* to be separated. All are rather irregular in shape, but in eggs of *T. axei* and *T. vitrinus* one end is usually pointed, while in those of *T. colubriformis* and *T. capricola* this character is seldom exhibited. The size differences of eggs of *Trichostrongylus* spp. are shown in Fig. 4.

SUMMARY

Fifteen genera of nematodes, the eggs of which occur in feces, have been reported in the United States as parasites of domestic ruminants. Eight of these genera have more than one species represented in one or more of the common domestic ruminants. Species of these genera may

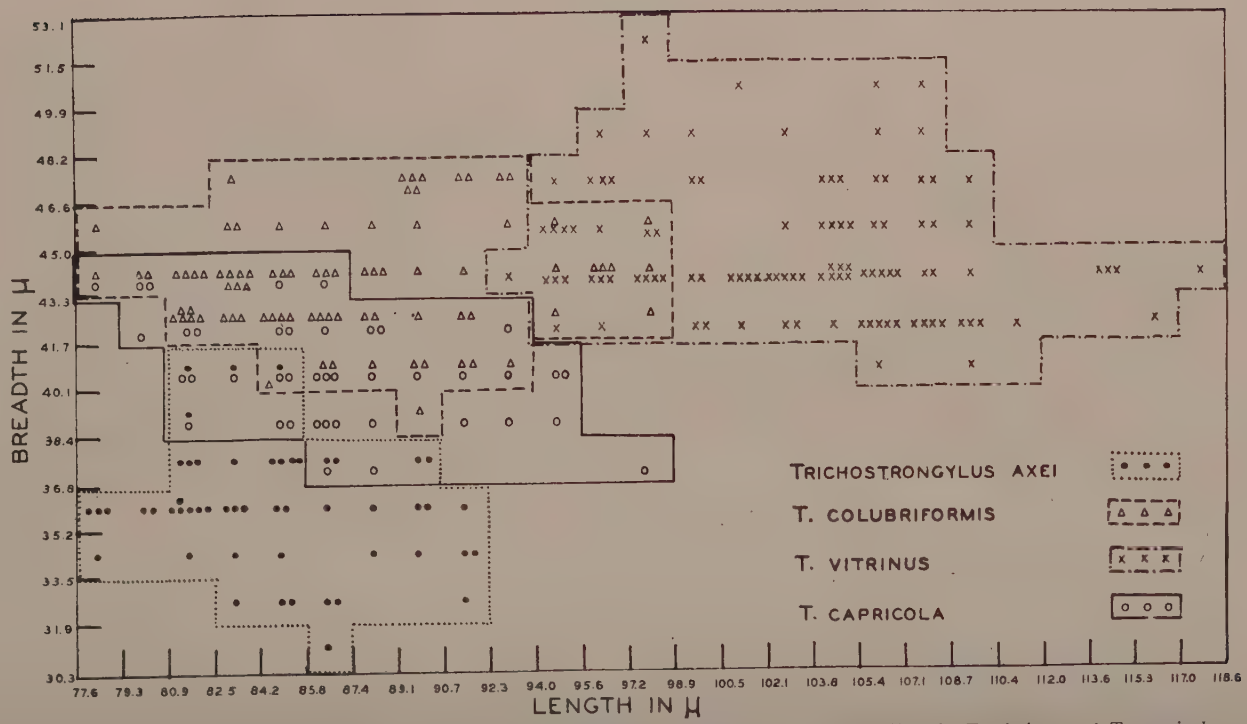

FIG. 4. Distribution of eggs within the indicated size ranges, *Trichostrongylus axei, T. colubriformis, T. vitrinus* and *T. capricola.*

be identified more or less definitely by differences in size and shape of their eggs. Precise identification is facilitated by host relationship and geographical distribution of the species considered.

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RESEARCH NOTES

THE EFFECT OF SULFAPYRIDINE ON EXPERIMENTAL AVIAN COCCIDIOSIS

In a previous report, (Levine, 1939, Cornell Vet., **29**: 309), it was shown that sulfanilamide, when administered to chickens by mixing it with the feed in a concentration of 0.3%, had a marked coccidiostatic effect (as measured by oöcyst counts) on five species of coccidia attacking the small intestine. A serious disadvantage in the feeding of this drug is that it manifests considerable toxicity for the birds as was shown by the fact that the weight gains of the treated birds at the end of the experiments were significantly less than those of the controls. Investigation of sulfanilamide derivatives was begun with the hope that a less toxic and perhaps a more effective compound might be found.

Since preliminary tests indicated that sulfapyridine in a concentration of 0.7% of the feed had a marked coccidiostatic effect, a trial was run to test its toxicity for growing chickens. Twenty birds, 7 weeks old, were divided into two equally-matched groups of 10 each. Sulfapyridine (0.7% in the mash) was fed to one group while the other received regular mash. Unfortunately, during the course of the experiment, three pairs of birds had to be eliminated because of miscellaneous conditions. After five weeks the average weight of the controls was 1.8 ounces greater than that of the treated birds. When this difference was analyzed statistically, however, it was found not to be significant. It was noted that when sulfanilamide was fed, the feces were wet and not well formed, indicating mild purgation. The fecal discharges of the sulfapyridine-fed birds, however, had the same appearance and consistency as those of the controls.

Three experiments were conducted to test the effect of sulfapyridine on coccidiosis. The birds in three groups of 10, 11, and 8 evenly-matched pairs (10–17 weeks old) were infected with equal numbers of sporulated oöcysts of *E. praecox, E. mitis,* and *E. hagani,* respectively. One group in each experiment received 0.7% sulfapyridine in the mash for three days prior to the infective feeding and for the duration of the experiment. The control group received non-medicated mash. The birds were kept in sterilized batteries with wire screen floors. Feed and water hoppers were so located that gross fecal contamination was impossible. The feces were collected from the dropping pans every day for oöcyst counts. The birds were handled and the oöcysts counted, as in the sulfanilamide experiments which were described in the article referred to above.

The figures in Table 1 represent the total number of oöcysts discharged by each bird daily. The controls infected with *E. praecox, E. mitis,* and *E. hagani* attained peak counts that were 58, 43, and 10 times greater, respectively, than those of the treated birds. If the total number of oöcysts passed by each bird during the first 8 days of the experiment is calculated, it will be seen that the controls passed 70, 89, and 10 times as many oöcysts as the treated birds. When the medicated feed was withdrawn from the treated groups, a gradual increase in the oöcyst discharge occurred and peaks were reached five days later. In no case were the peaks as high as those of the controls. The results obtained were practically identical with those obtained in the experiments with sulfanilamide.

One group of 10 birds was fed 0.7% sulfapyridine in the mash for two days. Five birds were then dosed with equal numbers of oöcysts of *E. tenella* while the other five were dosed with *E. necatrix.* A matched control group fed regular mash was infected with equal numbers of oöcysts of the same cultures. Three controls and three treated birds died of acute infection with *E. tenella* and two birds each from the control and treated groups of *E. necatrix* infection. There were no differences between the severity of the lesions in the treated and control survivors when they were sacrificed on the eighth day. Against these species, therefore, the

TABLE 1.—*Effect on oöcyst discharge of chickens through administration of 0.7% sulfapyridine in feed. The numbers represent the daily average, in hundreds of thousands of oöcysts, discharged per bird, after first appearance of oöcysts in feces.*

	Days on experiment	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
E. praecox	10 birds Control	4	776	1111	538	629	165	1.3	0.2	0.1	1.1	0.1	1.2	0.7	0.1	**	**			
	10 birds Treated	**	2	9	19	15	2	0.2	0.1	0.3	0.1	+ **	**	**	0.3	28	175	61	5.4	1.5
E. mitis	11 birds Control	3	67	86	101	97	74	28	28	20	0.4	0.3	0.5	0.4	0.5	0.1	**			
	11 birds Treated	**	1	1	0.6	2	0.4	0.1	0.1	0.7	**	+ **	**	**	1.0	5.3	50	12	2.4	0.8
E. hagani	8 birds Control	164	320	311	243	79	20	8	4	3	1	1	2	1	1	0.4	0.4			
	8 birds Treated	9	31	28	22	10	0.9	0.4	+ **	**	**	0.1	3.1	37.1	36.2	1.8	0.1			

+ Birds from this time fed non-medicated mash.
** Less than 7000.

action of sulfapyridine was disappointing. This had previously proven to be the case with sulfanilamide.

The data in this report indicate that sulfapyridine, like sulfanilamide, is an effective chemo-therapeutic agent against several species of coccidia affecting chickens. Sulfapyridine has an advantage over sulfanilamide in that it is non-toxic to the bird in effective concentrations. Unfortunately, neither of these substances is effective against the most destructive species, i.e., *E. tenella* and *E. necatrix,* and both are too expensive for practical use. The experience indicates, however, that the search for a chemo-therapeutic agent for coccidiosis is not a futile one, and that additional experimentation in this area is worth while.—P. P. Levine, *New York State Veterinary College, Ithaca, New York.*

THE EFFECT OF SULFANILAMIDE ON SEVERAL PARASITIC INFECTIONS OF LABORATORY RATS AND MICE

Sulfanilamide and other related compounds have in recent years produced remarkable therapeutic effects in a number of bacterial diseases. When applied to several infections with the animal parasites, the drugs have given variable results. Thus far substances of the sulfonamide type appear to have some value in the treatment of monkey malaria (Coggeshall, 1938, Am. J. Trop. Med. **18**: 715), and in coccidiosis of chicks (Levine, 1939, J. Parasitol., Dec. Suppl. **25**: 35) although in experimental trypanosomiasis (Kolmer and Rule, 1939, Proc. Soc. Exper. Biol. and Med. **40**: 77) and trichiniasis (McCoy, 1938, Proc. Soc. Exp. Biol. and Med. **38**: 460) the therapeutic results have been less favorable.

The present paper offers data obtained from a study of the effect of sulfanilamide, (1) upon experimental infection with a number of species of trypanosomes in rats and mice, and (2) upon natural infections with several species of intestinal protozoa in mice.

I. *The Effect of Sulfanilamide upon Experimental Trypanosome Infections. Method.* Experiments were carried out dealing with each of four species of trypanosomes: *Trypanosoma lewisi, T. cruzi,* and *T. equiperdum* in rats, and *T. duttoni* in mice. Infection was produced by injecting the animals intraperitoneally with a small amount (about 0.2 cc) of a broth suspension of trypanosome-infected blood. One-half of the animals used in each experiment were kept for control purposes; sulfanilamide was administered to the other half beginning on the day of infection and continuing daily thereafter. In the case of the mice, the drug was injected intraperitoneally; to the rats the drug was administered by mouth in 5% suspension. A uniform dose of 100 mgm of drug per 150 gm of body weight was given by either route. In addition, the treated animals received 0.5% solution of the drug to drink. The control animals were given no drug at any time and were supplied with water to drink. At regular intervals which differed with the different species of parasites, parasite counts were made in all animals. When the parasites exceeded 15 or 20 per high-dry field of the microscope, the counts were estimated by hemacytometer; with fewer parasites the actual number per field or, with *T. cruzi,* per 50 fields, was recorded.

Twenty-six rats were included in the experiment with *T. lewisi,* 24 in that with *T. cruzi* and 10 in that with *T. equiperdum.* Fifteen mice were employed in the experiment with *T. duttoni.*

Results: No animal was cured of its trypanosome infection by the administration of sulfanilamide, and in none was the course of the infection significantly altered favorably, compared with the controls. In some instances, particularly with *T. lewisi* and *T. cruzi,* the administration of the drug appeared to permit a somewhat more intense infection with the parasite, perhaps because of a deleterious influence of the drug upon the mechanism of resistance of the host. In the *T. equiperdum* infection, all ten of the animals used died of their infections within a few hours of each other on the fourth day after the parasite had been injected. This species was the only lethal trypanosome employed.

II. *The Effect of Sulfanilamide upon Natural Protozoan and Helminth Parasites of Mice.* The second part of the investigation was undertaken to determine the effect of sulfanilamide upon the presence of several protozoan parasites found naturally in mice. Four common parasites of mice were considered: *Endamoeba muris, Giardia muris, Trichomonas muris,* and *Hexamita muris.*

Method and results: Mice were treated with sulfanilamide (1) by feeding the drug with drinking water, or (2) by supplementing the fed drug with intraperitoneal administration.

In the first experiment, forty mice were used, 20 of these receiving 0.5% sulfanilamide solution to drink, and 20 receiving water to drink. After 25 days, all of the surviving mice were killed and the contents of their intestine searched for the presence of the 4 species of parasites selected for this study. By dividing the total volume of sulfanilamide drinking fluid used each day by the number of mice drinking the fluid, each treated mouse was estimated to have ingested about 25 mgm of the drug daily. Continued ingestion of the drug appeared to be toxic to the animals, for of 20 treated mice at the beginning of the experiment, only 8 survived until the day of autopsy. Nineteen of the 20 controls survived till autopsied. The results of this experiment failed to reveal any significant difference in the parasite incidence between the treated mice and the control group. The percentage incidence at autopsy for each of the four protozoa in the experimental and control groups was, respectively: *Endamoeba muris,* 50 and 55; *Giardia muris,* 87 and 77; *Trichomonas muris,* 50 and 77; and *Hexamita muris,* 87 and 61.

In the second experiment, 40 mice were used, 20 of these again being supplied with 0.5 per cent sulfanilamide solution to drink, and 20 being given water to drink. In addition, the former group was also injected intraperitoneally with daily doses of 10 mgm of the drug. After 10 days, all the mice were autopsied, and their intestinal contents searched for the four selected parasites. This time 19 of the 20 treated mice survived until the day of autopsy; all of the 20 control mice were alive until that day. The results again indicated that administration of the drug produced no significant effect upon the parasite incidence. The percentage of treated and control animals, respectively, infected with each species was: *Endamoeba muris,* 65 and 80; *Giardia muris,* 65 and 55; *Trichomonas muris,* 60 and 70; and *Hexamita muris,* 94 and 75.

Conclusion

Sulfanilamide was without effect in experimental infections with *Trypanosoma lewisi, T. cruzi,* and *T. equiperdum* in rats and in experimental infection with *T. duttoni* in mice. The drug was likewise without effect upon natural protozoan infections of mice with *Endamoeba muris, Trichomonas muris, Giardia muris,* and *Hexamita muris.*—Sophia Zuretт and James T. Culbertson, *Department of Bacteriology, College of Physicians and Surgeons, Columbia University, New York.* (This study was supported by a grant from the Committee on Therapeutic Research of the Council on Pharmacy and Chemistry of the American Medical Association. The sulfanilamide was generously supplied by the Winthrop Chemical Co., New York.)

ON THE ORIENTATION OF OPALINIDS

The fibrillar system of opalinid ciliate protozoans is completely asymmetrical; it, therefore, provides a means of orientation and comparison within the family Opalinidae.

Légèr and Duboscq (1904, Arch. Zool. Expér. et Gén. Ser. 4, **2**: 337) describing an anterior "zone arquée" in *Protoopalina saturnalis,* and van Overbeek de Meyer (1929, Arch. Protistenk. **66**: 207) a "Wachstumszone" in *Opalina ranarum,* indicate the presence of an anterior coördinating structure. Chatton and Brachon (1935, Compt. Rend. Soc. Biol. **118**: 958) describe a fibril, the "cinétie génératrice," from which longitudinal fibrils arch to and along the flattened surfaces of

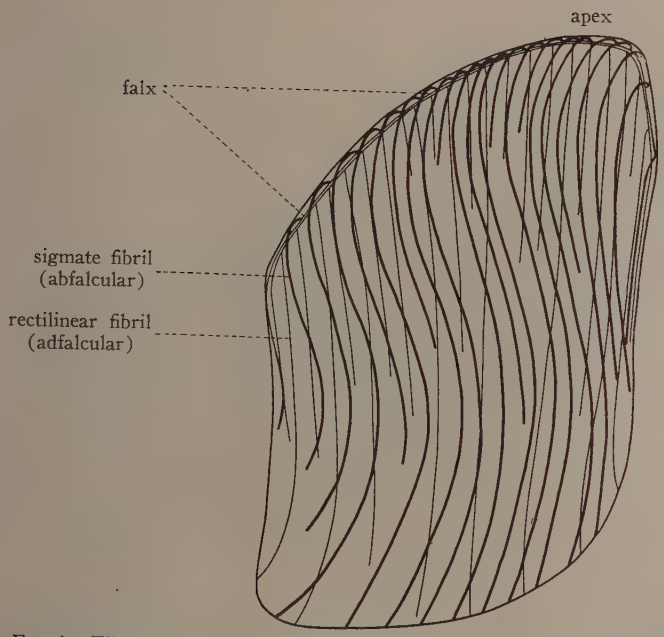

FIG. 1.—Fibrillar system of young individual of *Zelleriella intermedia*.

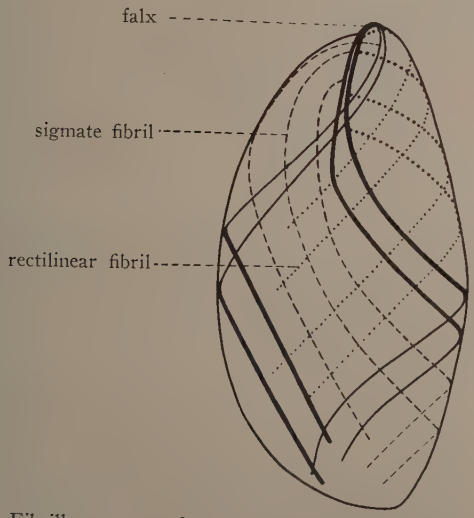

FIG. 2. Fibrillar system of cepedean larva of *Opalina draytonii*.

Opalina ranarum. Hara (1934, Botany and Zoology **2**: 2015; 1936, **4**: 537; 1937, **5**: 937; 1938, **6**: 1717) working on Japanese opalinids, determined that the "cinétie génératrice" is in fact a pair of fibrils. He states that they encircle a cytostome.

In *Zelleriella intermedia,* a binucleate, flattened opalinid from the rectum of a Louisianan toad, *Bufo valliceps,* the surface fibrillar system has two component groups of fibrils: the falx and the longitudinal fibrils. The *falx* is anterior and subterminal comprising two closely apposed falcular fibrils. The flattened surface on which the falx lies is *adfalcular* while the opposite surface is *abfalcular.* For convenience, but with no fundamental morphological significance, the adfalcular surface may be treated as ventral and the abfalcular surface as dorsal. In terms of these surfaces the *apex* or most anterior portion of the body lies to the right. Basal granules of cilia lie along the falcular fibrils; falcular cross-connective fibrils join some adjacent basal granules of the two falcular fibrils.

There are two sets of longitudinal fibrils rising from basal granules of the falx. Adfalcular or *rectilinear* fibrils rise from the granules of the posterior falcular fibril extending posteriad along the ventral surface in nearly straight lines. Abfalcular or *sigmate* fibrils rise from granules of the anterior falcular fibril to extend around the anterior end of the body, dorsad, and finally posteriad describing a narrow "S." An exception are a few fibrils rising from the apical end of the anterior fibril which are narrowly uncinate.

The fibrillar relations described for *Zelleriella intermedia* characterize other species of *Zelleriella* and *Opalina* examined. In *Protoopalina* and in cepedean forms of *Opalina* the falx is subterminal. Because the body form is cylindrical, there are no clearly defined dorsal and ventral surfaces. However, adfalcular fibrils may be recognized as similar to the rectilinear fibrils and abfalcular fibrils as related to the sigmate fibrils of flattened opalinids. All these types of fibrils may be demonstrated within the infection cysts of opalinids. Their fate in gamogeny and in copulation has not been discovered.

Because species have been described partly on the basis of the angle formed by longitudinal fibrils and falx, it must be noted that there are two such angles, of different size, in each individual opalinid.

Physiologically the "left" end of the falx or side opposite the apex is dominant. In normal forward motion ciliary waves originate here. In abnormal multinucleate zellerielles, mitosis has been seen to begin first at this dominant side and then in sequence in the nuclei toward the apex.—J. L. Mohr, *University of California, Berkeley.*

GLUGEA CAULLERYI NOM. N. FOR *GLUGEA MICROSPORA* VAN DEN BERGHE, 1939, PREOCCUPIED

In an abstract (1939, J. Parasitol. **25** Suppl: 21) I described a new sporozoan parasite from *Amnodytes lanceolatus* as *Glugea microspora.* Dr. Kudo has pointed out that the name is preoccupied by *G. microspora* Thélohan, 1892, a species considered by Kudo (1924, Illinois Biol. Monogr.) as identical with *G. anomala* (Moniez, 1887). Since the name *G. microspora* is not available, I propose for the new species from *A. lanceolatus,* the specific name *Glugea caulleryi.*—Louis Van den Berghe.

REVIEWS

NEMATODES PARASITIC IN ANIMALS. By Geoffrey Lapage, M.A., M.D. x+172 pages. Methuen and Co., Ltd., London. 1937. 4s. 6d. net.

This book appears to the reviewer to be an excellent critical review of the literature on the subject up to 1937. Dr. Lapage although not a helminthologist has during the past few years contributed some important facts and stimulating viewpoints to the study of nematodes. Four hundred and four titles were consulted and many of these are discussed by an active mind keenly interested in general biological problems.

Chapter I deals with structure, life histories, and classification. Wherever possible an attempt has been made to relate morphological structure to chemical composition and to function. Simple and complicated life histories are discussed and the many interesting variations in these emphasized. It is well for the student to beware of biological dogmas which are likely to explode at any time: "When any part of the life history is not parasitic, usually only the eggs and first and second larvae occur outside the host; but Potts refers to the peculiar reversal of this rule by species of *Mermis*, the sexual stages of which are all free-living in soil or water, the larvae being parasitic in insect larvae." A number of slightly disturbing inconsistencies occur. For instance on page 23 Lapage states that "Most nematodes are parasitic." Later, however, on the same page, after discussing the number of parasitic species, he states that "the number of free living genera and species is much greater." If the reviewer is not mistaken this last expression would have more nearly pleased the late Dr. N. A. Cobb. At the end of the first chapter, the author presents a skeleton-classification of species mentioned in the text.

The second chapter, entitled "Physiology," is the longest one in the book and from the viewpoint of the investigator perhaps the most stimulating portion, because really so little is known. Lapage feels that one reason for our ignorance is lack of interest in attempts to cultivate parasitic nematodes free from bacteria in artificial media, with which the reviewer is in emphatic agreement. This chapter deals with what is known of the nutrition of nematodes and its effects on the host; more specifically with the mechanical effects due to feeding on tissues or blocking of ducts, utilization of food or other materials required by the host, hemolytic, histolytic, anticoagulant and digestive secretions to which toxic properties are ascribed. The effects of migratory larvae are discussed: those which penetrate the skin or wall of the alimentary tract or cause pneumonia and other respiratory afflictions. The introduction of bacteria and protozoa or the assistance given to the entry of these through abrasions caused by nematodes are described, as in the work of Graybill and Smith on *Heterakis gallinae* and *Histomonas meleagridis*.

The work of McCoy, clarifying the earlier observations of Looss, on the food of the free-living larvae of hookworm, is described. On the other hand, the food of parasitic larvae and adult nematodes is not accurately known, and Lapage again rightly blames this on the inability to cultivate these stages. The ingestion of serum or tissue fluids and the entrance of nutrients by osmosis through a thinned cuticle adapted to this purpose as in the Filaroidea is outlined. Based on the work of Wells and others, Lapage states: "It can no longer be doubted that a great many species living in the alimentary canal take in blood in varying quantities," but he points out that no evidence exists that any of the blood-eating species digest the blood taken in. Whether materials in the plasma are used is not known. Hoeppli's work seems to suggest that *Ascaris lumbricoides* feeds on the intestine itself. This also seems to be the case with a spirurid, *Physaloptera clausa*, which ingests tissue histolysed around its head end. Very little appears to be known about the digestive secretions of nematodes although some evidence exists that *Ascaris* produces proteolytic enzymes and also has the ability to secrete antipepsin and antitrypsin with which to antagonize the pepsin and trypsin of the host.

The anemias associated with intestinal nematode infections are considered and ascribed to chronic loss of blood rather than to toxins or to hemolytic substances. "No one has shown that nematodes pass these substances into their hosts. If they did, they would be badly adapted."

Lapage describes McCoy's work indicating that the oxygen requirements of the free-living larvae of *Ancylostoma caninum* are low, which Brown had earlier shown true of the homologous stages of the pig ascaris. Species of *Ascaris* can take up oxygen and give off carbon dioxide even outside their hosts. The view also exists that *Ascaris* lives anaerobically and that glycogen present in the muscles, hypodermis, ovarian epithelium, esophagus and intestinal cells, is the chief source of energy. In discussing the possible formation of fatty acids by nemas the author believes "that the products of nematode metabolism cannot be accurately determined until a completely sterile nematode can be studied" because many of the products ascribed to nematodes may be produced by bacteria. Lapage supports the idea that blood-sucking forms like *Ancylostoma duodenale, A. caninum,* and *Haemonchus contortus* seek blood for its oxygen content rather than for any food constituents. It must not be forgotten, however, that in Wells's experiment the blood which passed out rapidly through the anus of *Ancylostoma caninum* was bright red arterial blood; not dark and reduced.

No accurate knowledge exists of the protein or fat metabolism by either parasitic or free-living nematodes. There is also no sound information concerning the functions of any of the so-called excretory organs, notwithstanding that excretion is usually ascribed to the lateral canal system and to the ventral gland.

Chapter III deals with the resistance of the host to nematode infections. Sandground is quoted on natural resistance which he regards as "an inherited character, an inherent incompatibility developed by adaptation during evolution." Lapage indicates that the problem of host specificity is complicated, however, "by the discovery of physiological strains within certain species which are morphologically identical, yet can infest different hosts and are, some authors think, restricted to these hosts." Scott described at least two strains of *Ancylostoma caninum,* and it is well known now that *Ascaris lumbricoides* shows two physiological strains. These strains are apparently instances of the selection of genetic races according to Chandler. Resistance of the individuals of a species of host to a particular species of nematode may also vary and undoubtedly this also has a genetic basis.

Age resistance must also be considered and has been demonstrated by many workers. The nature of age resistance, however, is not understood and the author thinks that "age resistance cannot yet be clearly differentiated from natural and acquired resistance." Lapage states that McCoy injected into rats antigens derived from nematode substance and presented the only evidence for the artificial production of acquired resistance. It appears that this author "produced resistance in rats and also in a proportion of monkeys a partial resistance, sometimes complete in the rats by injecting into them the substance of dead *Trichinella* larvae; but injection of the living larvae more effectively produced the same result. . . . Acquired resistance resulting from previous infestations with nematodes, whether these infestations have died out or still persist, is now well known and it is often an important protection to the host." Stoll, in his experimental work on sheep infected with *Haemonchus contortus,* "was one of the first to observe a self-cure of this kind." Acquired resistance to nematodes, therefore, is a reality but it usually develops slowly, requiring weeks or months. In regard to induced passive resistance Lapage mentions Chandler's view that there was not "any evidence that induced passive resistance to nematode infestations can be conferred by injection of the serum of hosts resistant to these parasites." Since 1937, however, Sarles and Taliaferro and Sarles have presented evidence that *Nippostrongylus muris* immune serum when transferred to non-immune rats confers resistance which has to a large extent an antibody basis.

Under factors that may lower resistance to nematode infections Lapage lists deficient diet, or any factor that lowers the general vitality of the host. He further states that a knowledge of factors that control resistance to nematodes is dependent upon more information on the metabolism of the parasites.

The fourth, i.e., the last chapter is the shortest one in the book and treats of the control of nematode infections. Here the failure to control many parasites of animals is emphasized and the opinion is again expressed that this failure must surely be due to our almost complete lack of reliable knowledge of the physiology of these

forms. In other words, "We know next to nothing about the factors which determine the success of their parasitic life, so that efforts to make that life impossible are without a rational basis." Nevertheless, in some cases successes have been attained through certain farm practices; moving animals frequently from pasture to pasture, plowing, chemical treatment of soil, destruction of intermediate hosts (snails), burning manure, etc.; also by disturbing the normal physiology of the parasites by the use of anthelmintics and finally by maintaining the resistance or tolerance of the host through suitable diet and general management directed to maintain health.

Lapage's book will remain a valuable and stimulating reference work for a long time, at least until most of the problems enumerated have been solved. The author deserves the gratitude of everyone concerned with the biology of living creatures.—R. W. GLASER.

MEDICAL ENTOMOLOGY. A survey of insects and allied forms which affect the health of man and animals. By William A. Riley and Oskar A. Johannsen. 483 pages, 184 illustrations. McGraw-Hill Book Co., New York and London. 1938. 2nd edition. $4.50.

The second edition of this useful textbook does not differ essentially in arrangement or mode of presentation from the first (reviewed in J. Parasitol. 1933, 19: 236). Nevertheless much of the new material which has accumulated in the field of medical entomology is mentioned, although so briefly that the second edition is only 7 pages longer than the first. Valuable keys are provided for the identification of the more common and important arthropods of medical importance. Also of great practical use are the sections devoted to methods for the control of such arthropods. The space used for such matters as the relation of cockroaches to cancer might perhaps have been put to better use. For example, there is no description of the internal anatomy of ticks, and no mention is made of some of the recent interesting and important contributions concerning the physiology of disease-transmitting arthropods. The illustrations are much the same as in the first edition. A supplementary bibliography covering 6 pages has been added which includes references to work appearing since the publication of the first edition. Unfortunately, several references cited in the text, as Syverton and Berry 1936, could not be found in either bibliography.—WILLIAM TRAGER.

MEDICAL ENTOMOLOGY. With special reference to the health and well-being of man and animals. By William B. Herms. 582 pages, 196 illustrations. The Macmillan Co., New York. 1939. 3rd edition. $5.50.

This book is a "completely revised and reset" edition of "Medical and Veterinary Entomology" (originally published in 1915) and is about 100 pages longer than the previous edition (of 1923, reviewed in this JOURNAL 1924, 11: 55). Although the title has been shortened, fully as much attention is paid to the veterinary as to the medical aspects. Following an historical introduction are chapters on the scope of medical entomology and methods employed in its study, on parasites and parasitism, on the general methods by which arthropods cause and carry disease, on the structure, development and classification of insects and arachnids, and on insect mouth parts. The succeeding sixteen chapters are devoted to a consideration in taxonomic order of the various groups of insects and arachnids of medical and veterinary interest, and keys are provided for their identification. The last two chapters deal with venomous and urticarial arthropods and with the use of arthropods in medical practice. Each chapter is followed by appropriate bibliography, and each subject is discussed in a very thorough and satisfactory manner. The taxonomy, morphology, life history and control of the more important species are adequately treated, so that the book is excellent not only as a text but as a reference for workers both in the experimental and practical fields of medical entomology. In view of the importance of the subject, a more extended discussion of the different races of *Anopheles maculipennis* and their relation to malaria would

have been desirable. There is a tendency to devote a slightly disproportionate amount of space to work in which the author and his immediate associates have participated, but this is a not wholly undesirable characteristic. The value of the book is greatly enhanced by the numerous illustrations. Most of these are from original photographs and drawings and are beautifully clear and accurate. For example, the plate (p. 164) illustrating the life histories of mosquitoes is a model of concise pictorial presentation.—WILLIAM TRAGER.

GERMAN-ENGLISH SCIENCE DICTIONARY FOR STUDENTS IN THE AGRICULTURAL, BIOLOGICAL AND PHYSICAL SCIENCES. By Louis De Vries. x+473 pages. McGraw-Hill Book Co., New York. 1939. $3.00 postpaid.

The author, with the collaboration of 34 colleagues on the graduate faculty at Iowa State College, has produced an exceedingly useful and usable dictionary. The book is of convenient size, bound in a semi-limp cover and printed with double column pages. The boldface Roman type used for the German words and root stems provides a good contrast to the English equivalents which are in standard Roman type. Although the type size is small (six on seven point), the use of four to six point leads between each boldface German heading makes the words and lines stand out distinctly. This clarity makes for a degree of ease in use seldom encountered in a dictionary, particularly in one dealing with a foreign language.

The scope of the work is ambitious. In the introduction the author indicates that this dictionary is the first of its kind, compiled especially for use by students in science. Subjected to somewhat extensive use by the reviewer, it fulfilled the requirements to a high, though not perfect, degree. In part this was due to the fact that parasitology and zoology were not as well represented in the field of interest of the collaborators as they might have been. The concentration of botanical interest finds partial reflection in the dictionary in what the reviewer considers to be an inordinately large number of common names of plants. From this point of view the volume lacks the balance of an ideal science dictionary. Numerous words are included which are orthographically and unmistakably identical with their English equivalents and seem conspicuously unnecessary. This leaves room for improvement in quality in subsequent editions (which the volume richly deserves) without proportionate increase in bulk.

The emphasis on short root stems which make up such a large proportion of technical German words is commendable and reduces to a minimum the possibility of erroneous deductions on the part of the user.

A surprisingly small number of typographical errors was encountered. That a few are present merely indicates that proof-reading is "noch nitch" [sic, p. 282] an easy task.—G. L. GRAHAM.